

GROWTH, REPRODUCTION AND SURVIVAL
OF SOME MARINE COPEPODS SUBJECTED TO THERMAL
AND MECHANICAL STRESS

By

RAYMOND WILLIAM ALDEN III

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Raymond William Alden III

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Chairman: Frank J. S. Maturo, Jr.
Major Department: Zoology

A study was made of the lethal and sublethal effects of power plant entrainment and thermal stress on the copepods of the Crystal River estuary on the west coast of Florida. The important copepod species that were observed for entrainment mortality listed in order of abundance were Oithona spp., Acartia tonsa, Paracalanus crassirostris, Euterpina acutifrons, Pseudodiaptomus coronatus, Tortanus setacaudatus and Labidocera spp. Experimental field treatments employing circulating system drift bottle devices were set up to test the relationships between entrainment mortality and such variables as temperature, salinity, length of exposure to the heated effluent, mechanical damage, seasonal factors, density (numbers/m³) and sex or age class of the various species. Temperature, salinity, and temperature-salinity interactions were seen to be among the most important factors influencing mortality. Under conditions of low temperature and moderate salinities, mechanical damage was the major lethal entrainment effect for most species, but produced relatively low mortalities compared to those found with other temperature-salinity

regimes. Each species had unique response patterns to temperature-salinity effects, as well as to combinations of other variables, but virtually all exhibited rapidly increasing mortalities as temperatures rose above 35°C.

Laboratory studies were carried out to observe the subtler biological aspects of entrainment and thermal stress: the effects on reproduction of copepods surviving entrainment; the effects on growth of entrained juveniles; and the long-term effects occurring over a period of time following entrainment. Sublethal entrainment effects were negligible until summer conditions caused discharge waters to rise above 35°C. Above this temperature, fecundity rates declined and growth rates of juveniles dropped below those of control populations. Long-term mortality rates of copepods surviving entrainment were not significantly different from those of controls, although the mortality rates of both populations were shown to be accelerated by increasing culture temperatures.

Possible biological and ecological implications of entrainment and thermal stress are discussed in light of the findings of the present study.

SECTION I

INTRODUCTION

The important role of copepods in the sea has been long recognized. These holoplankters dominate the zooplankton in most estuarine and marine environments and play the principal "middle man" in the transfer of energy from primary producers to higher trophic levels (Clarke and Gellis, 1935). The importance of these organisms is especially apparent in estuaries, where ecological adaptations allow certain species to utilize the energy from these nutrient-rich ecosystems to form blooms of vast numbers. Migrating species of higher trophic levels make efficient use of this food supply by utilizing estuaries as breeding and nursery areas. It has been repeatedly demonstrated that copepods play the major food item for most of the young and at least some adult fishes (Grice, 1957).

The vital role that copepods play in the functioning of estuarine ecosystems makes the knowledge of the biological responses of these organisms to environmental factors extremely important. Both drastic and subtle responses to environmental changes may affect the survival, distribution, and productivity that make the copepods so important to the system. The ability to predict the biological reaction of important copepod species to environmental stress, either natural or man-made, may be the first step in the effective management of food chains that support a sizable portion of the world's fisheries.

Planktonic organisms are subjected to many environmental stresses that may be actively avoided by the nekton as well as many benthic animals. It is for this reason that much interest has been focused on the resistance of the planktonic forms to such factors as thermal stress (Strickland, 1969). Recently, studies in this area have shown a trend away from the univariate analysis of temperature tolerance and towards a multiple factor approach. Interactions of temperature with such factors as salinity, dissolved oxygen and duration of exposure have been seen, under many conditions, to be as important in shaping the tolerance pattern of a species as the primary response to temperature (Alderdice, 1972). Laboratory studies have been performed in an attempt to define the effects of such interactions on the thermal tolerance levels of planktonic larvae of crustaceans (Costlow et al., 1960, 1962, 1966), molluscs (Kennedy et al., 1974; and Lough, 1975), and fishes (Alderdice, 1963; and Alderdice and Forrester, 1968). These studies have utilized multiple regression analysis and response surface techniques to define the tolerance patterns of the experimental organisms to various environmental factors. The methodology of such an approach has been reviewed and the biological applications discussed in detail by Alderdice (1972).

A related area of study, prompted by growing concern over industrial thermal pollution, is that of the effects of entrainment of zooplankton in the cooling systems of power generating plants. There are recent reviews of such entrainment studies by Raney (1973) and Coutant and Pfuderer (1974), as well as the proceedings of a symposium on the state of the art edited by Jensen (1974). Entrainment studies usually involve on-site field work, since exact conditions experienced

by organisms pumped through a power plant cannot be artificially reproduced under laboratory conditions. As a result of the limitation on experimental design, statistical treatment of entrainment mortality is generally simpler and less descriptive than that of laboratory thermal tolerance studies. Zooplankton mortality is usually related by linear regression analysis to the single independent variable of discharge temperature (Icanberry and Adams, 1974) or to the combination of discharge temperature and thermal rise (Davies and Jensen, 1974). Interactions of environmental factors and entrainment effects of the type described by response surface methodology, for the most part, have not been examined by entrainment studies.

Another area that is generally overlooked by entrainment studies is that of sublethal entrainment effects. Subtle post-entrainment changes in important processes such as growth, reproduction and long-term survival could have nearly as significant an effect on the zooplankton population as immediate mortality (Levin et al., 1972; and Strickland, 1969). Heinle (1969) studied the effects of entrainment on growth of copepods and reported a depression in growth rate for the populations going through the power plant. This indication of significant sublethal entrainment effects suggests that all biological processes that might affect the productivity of a species should be studied.

The present study was designed to examine lethal and sublethal effects of entrainment and thermal stress on estuarine copepods. The investigation can be divided into two basic sections: an experimental field study designed to analyze the environmental factors that determine the mortality suffered by entrained species of copepods, and a

laboratory study that was set up to observe subtle post-entrainment effects on various biological processes. Considered in the field study of entrainment mortality were such factors as intake and discharge temperature, salinity, mechanical damage, delayed thermal effects, seasonal effects, and density, sex and age class of each of the major species of copepods. These variables were analyzed to observe how various combinations of factors shaped the response patterns of the entrained organisms. The laboratory study supplemented this information by observing entrainment and thermal effects on the reproduction, growth and long-term survival of selected species of copepods. The study was designed not only to assess the impact of a specific power plant, but also to observe general biological responses of the species of copepods to thermal stress and the environmental factors that influence these patterns.

SECTION II

METHODS AND MATERIALS

Field Studies

Description of Field Site

Research was conducted at the Florida Power Corporation steam generating plant which is located between the Withlacoochee and Crystal Rivers on the west coast of Florida. This area represents the typical shallow water, middle salinity estuary found along the Gulf coast of the state. Water temperatures generally range annually from 14 to 30°C (Figure 1) and salinities range from 17 to 28 ppt. (Figure 2). Seasonal freshwater runoff from surrounding terrestrial and marsh areas during rainy seasons causes fluctuations in salinity, detrital content, and organic and inorganic constituents of the water. The spring-fed rivers of the area, however, provide a constant stabilizing force not found in many northern estuaries.

The two steam generating units now in operation at Crystal River have a combined electrical capacity of 897 megawatts and use up to 640,000 gallons of water per minute from the estuary for cooling purposes. The power plant raises the temperature of the cooling waters passing through it an average of 5.88°C (Figure 1). This thermal increase (ΔT) is quite constant because environmental laws prohibit ΔT values above 6.5°C and plant operation efficiencies prevent minimum ΔT values much below the mean value. Surface salinities are generally

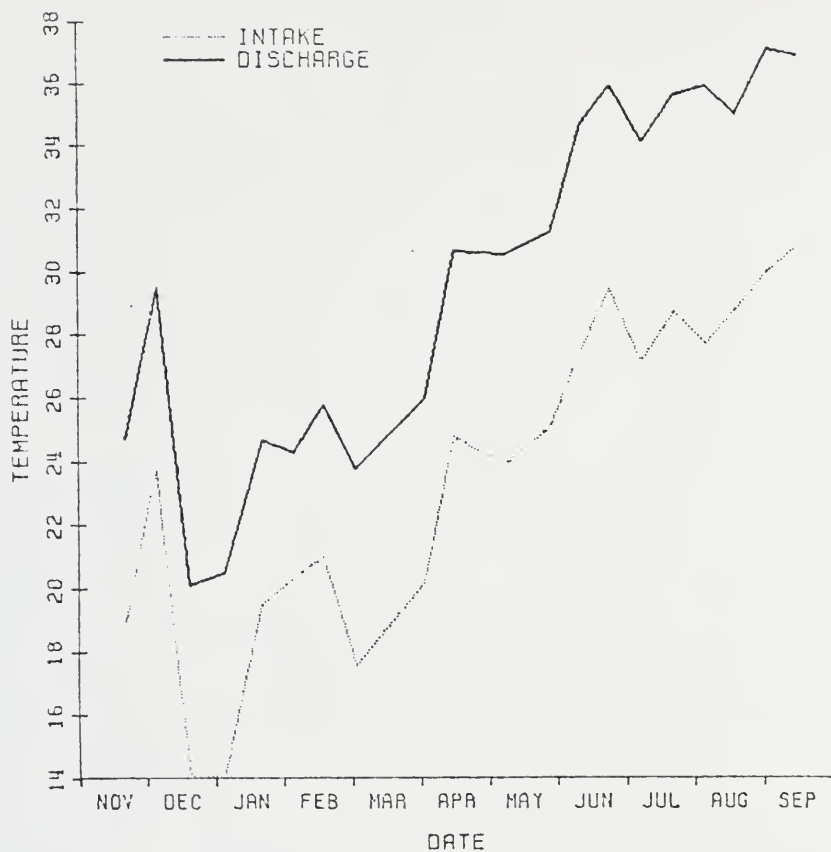


Fig. 1. Seasonal variation in water temperature in intake and discharge canals of the Crystal River power generating plant.

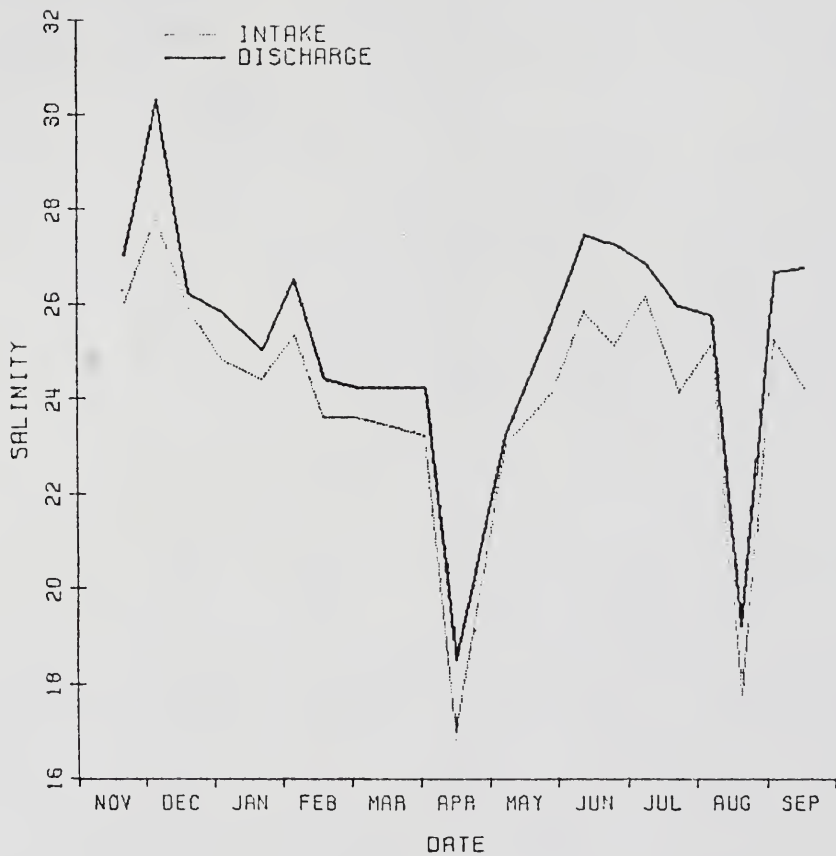


Fig. 2. Seasonal variation in salinity in intake and discharge canals of the Crystal River power generating plant.

raised by 1.12 ppt. (Figure 2) in going through the plant because the higher salinity oceanic water from the bottom is mixed with lower salinity surface waters when the cooling water is taken from the partially stratified estuary. The heated effluents are channeled away by a mile-long discharge canal, which takes the water exiting the plant approximately two hours to traverse during normal operations. During the passage of the discharge waters down this canal, temperatures remain relatively unchanged until they are released into the open bay area of the estuary.

A proposed nuclear-powered generating unit now under construction at the Crystal River site will approximately double the electrical capacity and the maximum flow of cooling water of the power plant. The new unit is designed to prevent increase in ΔT values, but will increase the area of thermal impact due to the larger volume of heated discharge waters. The numbers of planktonic animals entrained would also approximately double with the increased demand for cooling waters.

Experimental Design

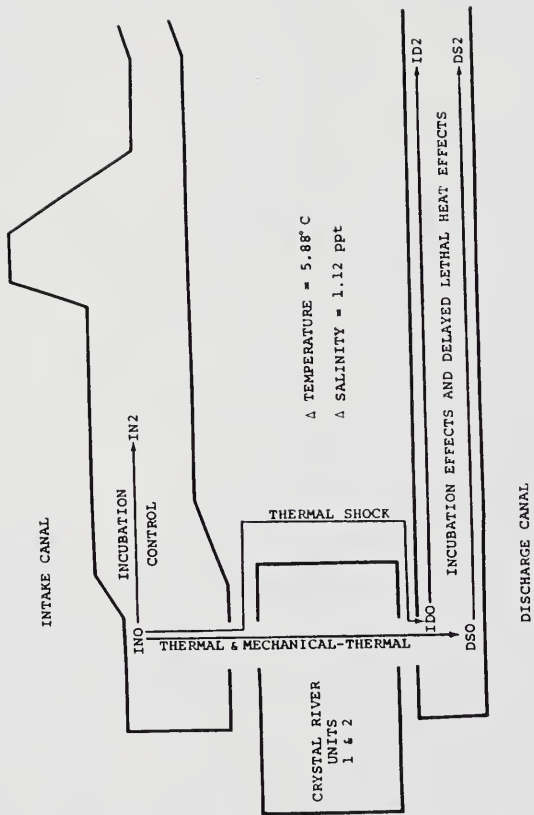
Copepods were collected utilizing a 64 micron mesh 0.5 meter plankton net fitted with a digital flow meter. Samples were taken biweekly from intake and discharge areas from November 1973 through September 1974. The number and length of net tows were adjusted so that approximately equal volumes were sampled from each area and so that the minimum amount of mortality was caused by collection techniques. A pump-net filtration system based on that described by Icanberry and Richardson (1973) was tested as a sampling device designed to reduce

collection mortality, but was shown statistically to be avoided by escape mechanisms of certain types of zooplankters (Maturó et al., unpublished data). Thus, the pump system was abandoned for net collection. Temperature and salinity measurements were taken with a Beckman salinometer at each area during every sampling period.

Six experimental field treatments were set up to attempt to separate various factors involved in entrainment mortality (Figure 3). Two controls were made utilizing copepod populations from the intake area. The initial control (IN0) is a population that has been collected and analyzed immediately for mortality by vital stain bioassay (Dressel et al., 1972). The second intake treatment (IN2) subjects an intake population to 2 hours of incubation in a circulating system drift bottle apparatus of the type described by Gonzalez (1973). This device is placed in the intake waters and the sample, which is treated for bioassay after the incubation, acts as a control for the delayed thermal exposure treatments described below.

A third intake population of copepods (ID0) is brought to the discharge area, placed in a screened PVC container and submerged in the heated effluent until it has been brought up to discharge temperatures (approximately 5 minutes). The sample is then treated for bioassay and the mortality above control (IN0) values are assumed to be due to the effects of immediate thermal shock. A discharge sample (DS0) is also assayed immediately for mortality, this population having undergone the turbulence, shearing forces and mechanical damage associated with passage through the condenser system of the power plant. Since the only difference between the ID0 and the DS0 populations is that the latter has been subjected to the mechanical effects of

Fig. 3. Schematic diagram of study site indicating designation and purpose of experimental field treatments. See text for explanation of abbreviations.



entrainment, the damage caused by the physical factors of plant passage can be assessed by regression analysis.

Delayed thermal effects associated with the amount of time that the population spends in the heated effluent of the discharge canal are also examined. An intake population (ID2) and a discharge population (DS2) are placed in a drift-bottle apparatus similar to that used in the IN2 treatment and are allowed to drift down the length of the discharge canal for 2 hours. These organisms experience an exposure to the heated canal waters similar to that experienced by a natural population going through the power plant. The ID2 population, however, has not experienced the mechanical effects of entrainment and might more closely represent a population entrained into the thermal plume by tidal recruitment. The results of these treatments allow the assessment of the delayed thermal effects of entrainment encountered within the thermal plume.

Samples from each field treatment were preserved in 5% formalin and kept cold to preserve stain intensity until counts were made (Dressel et al., 1972). Immediately before counting, the samples were sieve-fractionated into three size classes by pouring them through a series of geological sieves (300, 150 and 75 micron mesh) that were clamped to a "wrist-action" shaker. This method allowed each size class to be split down separately for counting with a Folsom Plankton Splitter so that the large, rarer animals were not "lost" in the splitting of a sample to countable size. The neutral red stain in the organisms was "fixed" by titration with an acetic acid-sodium acetate mixture (5N). This method of acidification to bring out the red coloration immediately prior to counting was found to be far superior

to that of using acidified formalin during preservation (Dressel et al., 1972) because the acid tends to cause the neutral red to leach from the bodies of the copepods, reducing the amount of time that the samples can be sorted before the stain fades to an undetectable intensity. The samples were counted under a dissecting microscope at 25X or 50X, depending on size class. The copepods were identified to species and sex and notations made as to whether they were alive or dead upon sampling.

Mortality values for the control populations (IN0, IN2) were assumed to be baseline levels and were thus subtracted from those of the populations that were subjected to the experimental field treatments (ID0 and DS0, ID2 and DS2) to obtain values for mortality caused by the various factors associated with entrainment. On those sampling dates that the experimental population of any given species reached 100%, entrainment-induced mortality was assumed to be 100% and control values were not subtracted. The entrainment mortality data for each species subjected to the various field treatments were analyzed by multiple regression analysis. Predictive regression models for entrainment mortality under various conditions were formed by the Stepwise MAXR Regression computer program from the Statistical Analysis System (SAS) contained in the CIRCA Computing Library at the University of Florida. The independent variables tested were temperature, salinity, delayed thermal, mechanical and seasonal effects, along with effects associated with the sex, age class and density (numbers/m³) of the species of copepod being observed. Fourth order temperature and salinity terms were included in the statistical model. Preliminary regressions indicated that the increase in the amount of

variation explained (R^2) ceased to be significant for terms beyond the fourth degree. All reasonable interactions between the independent variables were also tested. The stepwise regression program was of the type described by Lough (1975) and the predictive model selected from the sequence was the one that produced the greatest values of R^2 for the most parsimonious model. More complex models were observed to make sure no major increases in R^2 values occurred with the addition of various combinations of variables.

The regression equation for each species was used to plot response surfaces for various entrainment conditions utilizing the SYMVU three-dimensional plotter routine from the CIRCA Computing Library. Temperature and salinity, being the major physical factors, were plotted on the X and Y axes, while the predicted mortality values were plotted on the Z axis in the third dimension. All other factors were then varied one at a time to observe their effects on the response of the species for the salinity and temperature conditions. The predicted mortality values were rounded up to the nearest 10% level so that all trends would be clearly discernable on the graphed surfaces.

Laboratory Studies

Growth Studies

To determine the effects of entrainment on growth of juveniles, a method modified from that used by Heinle (1969) was employed. During the biweekly sampling period, juvenile Acartia tonsa and Oithona brevicornis, the two dominant copepods of the area, were collected for

the growth experiments. The waters from intake and discharge areas were bucketed through a 73 micron mesh 0.2 meter diameter plankton net that was clamped into the mouth of a large, perforated PVC container. The perforations in the PVC container were screened with 5 micron filter bag material and the entire apparatus was suspended vertically in the water during the sampling procedure. Approximately 80 gallons of water were filtered from each canal by this method, although high densities of juveniles during the summer months allowed the volume sampled to be cut in half. The 73 micron net filtered out all but the smaller naupliar stages, while the PVC container retained and concentrated these early instars.

The concentrated samples of juveniles were kept in insulated containers filled with sea water from the collection areas for transport back to the laboratory. In the laboratory, the intake and discharge samples were each split into eight equal subsamples with a Folsom Plankton Splitter that had been darkened to prevent phototrophic clumping on either side of the divider. The subsamples were placed in culture dishes containing 1 liter of fresh sea water to which had been added a mixed algal diet. The concentration of this food mixture was adjusted to give a final concentration in the cultures of 60,000 cells/ml.: 24,000 cells/ml. of Rhodomonas baltica; and 12,000 cells/ml. each of Isochrysis galbana, Monochrysis lutheri and Thallasiosira pseudonana. The algae were grown in pure cultures, utilizing Guillard's f/2 media (Guillard and Rhyther, 1962) and concentrations were determined by counting fixed cells from a serial dilution in a hemacytometer. A few drops of a culture of large marine ciliates (probably Euplotes sp.) were routinely added to the food culture before feeding. These

organisms help eliminate excess buildup of bacteria and algal detritus in the bottom of the culture dishes and provide a secondary food source for the older stages of the copepods (Zillioux, 1969). The developing juveniles were further protected from being entangled in this algal detritus layer by blackening the bottom of the culture dishes to cause phototrophic attraction away from the area where the debris settles.

One culture from each population was stained and preserved immediately, while the remaining cultures were kept in an illuminated B.O.D. box set at intake photoperiod and temperature conditions. Cultures from each population were then chosen randomly at predetermined time intervals, stained, and preserved. The time interval chosen depended upon the development time for the particular season: every other day in winter, and every day in summer when higher temperatures caused accelerated growth. Remaining cultures were fed every other day, with the mixed algal diet being added to bring the concentrations in the culture dishes up to proper levels.

The samples were counted under a dissecting microscope at 50X and the Acartia tonsa and Oithona brevicornis nauplii and copepodites were identified as to stage of development. The mean stage was then calculated for each day that a culture was sampled by the following equation:

$$\bar{N} = \frac{\sum_{i=1}^N N X_N}{\sum_{i=1}^N X_N}$$

\bar{N} is the mean stage for any given day, N is the stage number up to

the 12th or adult stage and X_N is the number of individuals counted in any stage N.

Growth curves for intake and discharge populations were graphed for each experiment by plotting mean stage versus day. Linear regression analysis of the curves produced "B" values or slopes which are the growth rates for each population of each species. These rates were then compared with a T-test designed to compare slopes. Thus, the growth rates of intake and discharge populations for each date were compared statistically to determine any significant differences due to the effects of entrainment.

The growth rates for all experiments were plotted against date to look for any visible trends in divergence among the populations. Regression analysis was performed to test the relationship of growth rate to the area from which the juveniles were collected, to the date on which they were collected and to the interaction of these factors.

Reproduction Studies

In order to determine the effects of entrainment on reproduction, intake and discharge populations of copepods were collected in a 202 micron plankton net and transported to the laboratory in insulated containers. Male and female Acartia tonsa were sorted out under a dissecting microscope, utilizing a large bore pipette with a mouth tube. Generally, females without attached spermatophores were selected in an attempt to start all cultures at the same point and to test male fertility. Two males and two females were placed in one-liter containers of fresh sea water and fed the mixed algal diet every other

day. The cultures were kept in a B.O.D. box set to simulate intake temperatures and photoperiod for 5 to 7 days. The populations were then prepared with vital stain and preserved for counting. The adults, copepodids, nauplii, and eggs from each container were counted under a dissecting microscope. An eggs/female-day ratio was then computed from the total reproductive products and the number of living females.

It became apparent that this experimental design was not very efficient, since all cultures in which a female had died during the culturing period had to be discarded as it was impossible to ascertain how many, if any, productive days there were before mortality. A change in method was therefore made and the cultures were set up as pair-mating experiments, with a single male and a single female Acartia tonsa being placed in half liter containers. These cultures were then observed daily for any mortality. When a female was found dead, the male was transferred to a fresh container and the eggs and juveniles were immediately stained and counted. In cultures where the male was found dead, it was replaced with a live male from the stock sample and note made as to the day on which the mortality occurred. To calculate the eggs/female-day ratio for these pair-mating experiments, the day number was assumed to be that of the day on which the female was last observed to be alive. Any discrepancies between the estimated productive time and the amount of productive time that the female might have actually had before the observation of mortality were assumed to average out between the populations.

Multiple regression analysis was used to test the relationship of the rate of egg production to the area from which the adults were taken, the date on which they were collected and the interaction of these

factors. A Duncan's Multiple Range test from the Statistical Analysis System (SAS) computer program package was performed to determine the dates on which the reproductive rate of the entrained population differed from that of the control.

Analysis of Long-Term Survival

The culturing of copepods for the growth and reproductive studies provided data for the observation and testing of differential survival between intake and discharge populations for a relatively long period of time following entrainment. The comparisons, although dealing with laboratory conditions, were assumed to show any drastic differences that may occur between the populations in nature.

The growth experiments provided estimates of the total number of juvenile Acartia tonsa and Oithona brevicornis alive on each day of observation. Such data allowed the plotting of survivorship curves of the juveniles from intake and discharge populations for each sampling period. The slopes of the curves were determined by linear regression analysis and compared by a T-test.

Data from the pair-mating experiments produced the daily observations of adult Acartia tonsa mortality during the 5 to 7 days following collection. Survivorship curves for the populations in each experiment were constructed, plotting the percentage of the adults remaining alive on each day. The slopes for these curves were also determined by linear regression and compared with a T-test.

As a second test of long-term adult survival, the total amount of mortality found during each reproductive experiment was used to

calculate an average percent mortality/day value for the period. Values calculated in this manner were rougher estimations of mortality rates than those obtained by the regressional fitting to daily data, but such calculations allowed inclusion of data which were collected before daily observations were established (December 1973 to April 1974). Regression analysis of mortality rates from the period from April to August when both estimations could be made showed no significant differences (at 0.05 level) between the methods, so it is assumed that the rough estimations adequately show trends in survival.

SECTION III

RESULTS

Field Studies

Acartia tonsa

Acartia tonsa seems to be the major year-round component of secondary production in the zooplankton community of the Crystal River area. The numbers of this species were exceeded only by those of the Oithona spp., which, because of their smaller size, probably do not contribute nearly as much to the biomass production of the area. The numbers of adult A. tonsa ranged from $10/\text{m}^3$ to $1,208/\text{m}^3$ for males and from $43/\text{m}^3$ to $1,835/\text{m}^3$ for females, with mean values of $283/\text{m}^3$ and $444/\text{m}^3$ respectively (Figure 4). Numbers of juvenile A. tonsa ranged from $1,127/\text{m}^3$ to $14,371/\text{m}^3$ with a mean of $5,795/\text{m}^3$. Peak numbers occurred in December, late February to early April, July and September. This species annually comprises 10 to 50% of the total copepod population with values of 30 to 40% for most of the year (Figure 5).

Acartia tonsa that were exposed to discharge temperatures and sampled immediately (treatment ID0) generally had low mortality values. Mortality for the species as a whole for the ID0 treatment ranged from 0 to 32%, with a mean value of 6.9%. Males exhibited higher mortalities than females, while juveniles generally showed lower mortalities

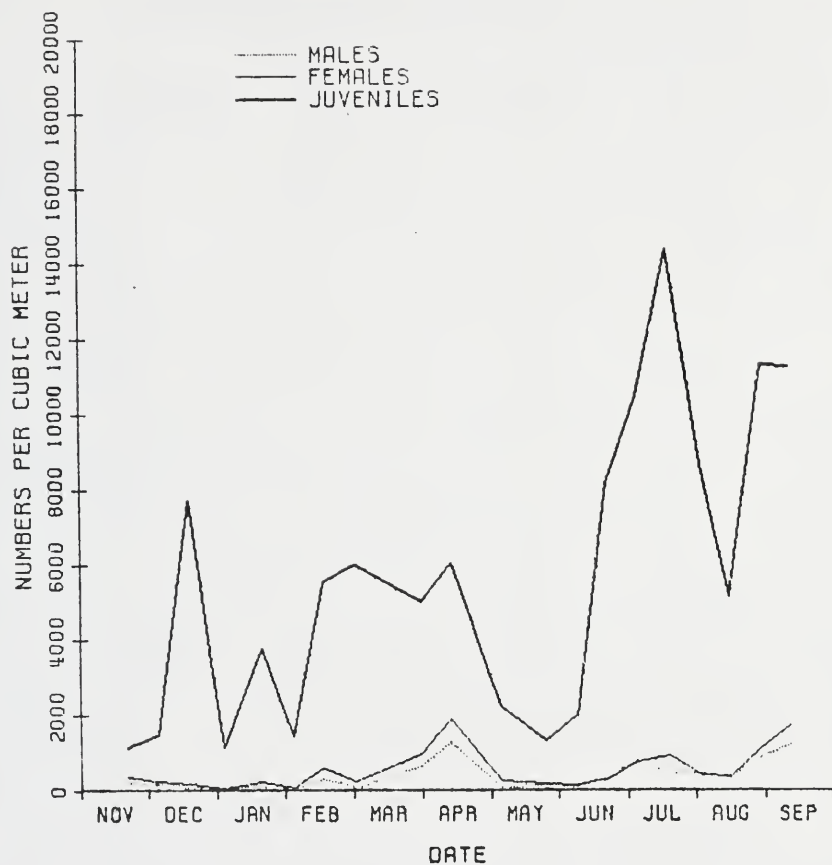


Fig. 4. Seasonal variation in numbers of *Acartia tonsa*.

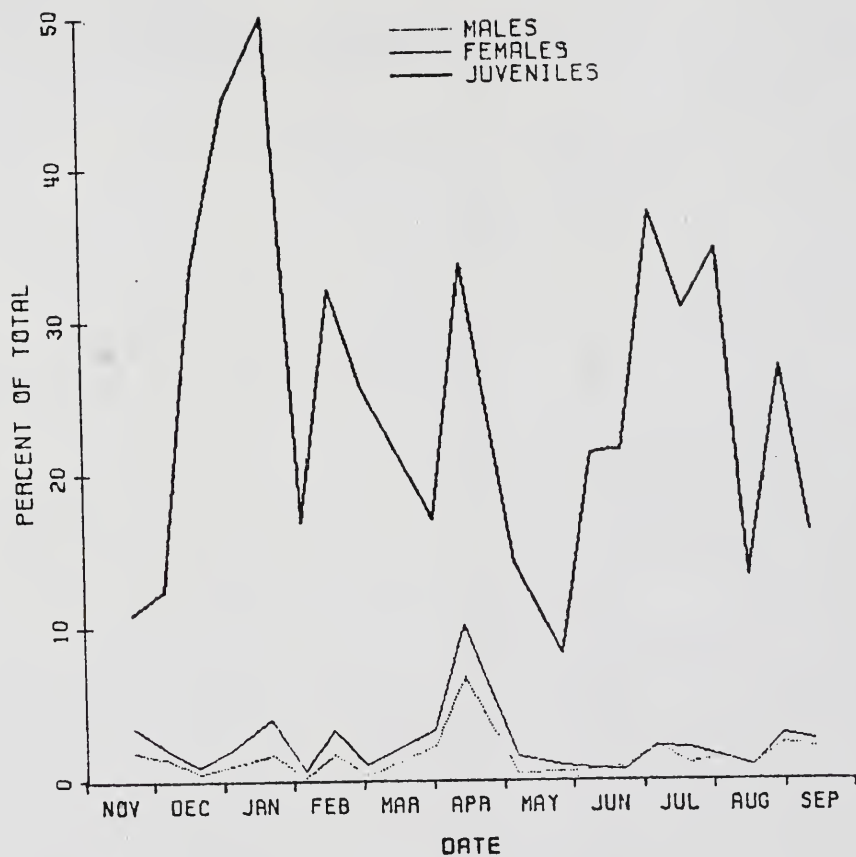


Fig. 5. Seasonal variation in percent of total copepod population represented by *Acartia tonsa*.

than either adult (Figure 6). There was a trend of low mortalities in the winter and generally higher mortalities in the warmer months.

Populations of Acartia tonsa that were exposed to thermal shock and then allowed to remain in the heated discharge water (treatment ID2) exhibited greater mortalities than those experiencing only the thermal shock. The mortality for the species as a whole ranged from 0 to 80% with a mean of 15.6%. Again, males tended to be the most sensitive to mortality, while juveniles were the least sensitive (Figure 7). The trend through the year at ID2 was for mortalities around 10% through most of the winter and spring months, with a rise through the latter part of the spring and a sharp increase in mid-summer.

The Acartia tonsa population that has been sampled immediately upon leaving the plant (treatment DS0) generally showed higher mortalities than those experiencing only the thermal shock (ID0). The range of mortality values for the species as a whole exposed to treatment DS0 was from 5 to 40% with a mean value of 12.4%. The males had almost consistently higher mortalities than females, while the juvenile values were nearly always lower (Figure 8). Mortalities were moderately low throughout the year, with a slight trend to higher values in the summer.

Acartia tonsa that were entrained through the power plant and then allowed to drift down the discharge canal (treatment DS2) exhibited the largest mortality values. Mortalities for the population as a whole ranged from 5 to 80% and had a mean of 16.7%. A clear trend can be seen, with mortalities being moderately low throughout the winter and spring, gradually rising through the early summer and then sharply

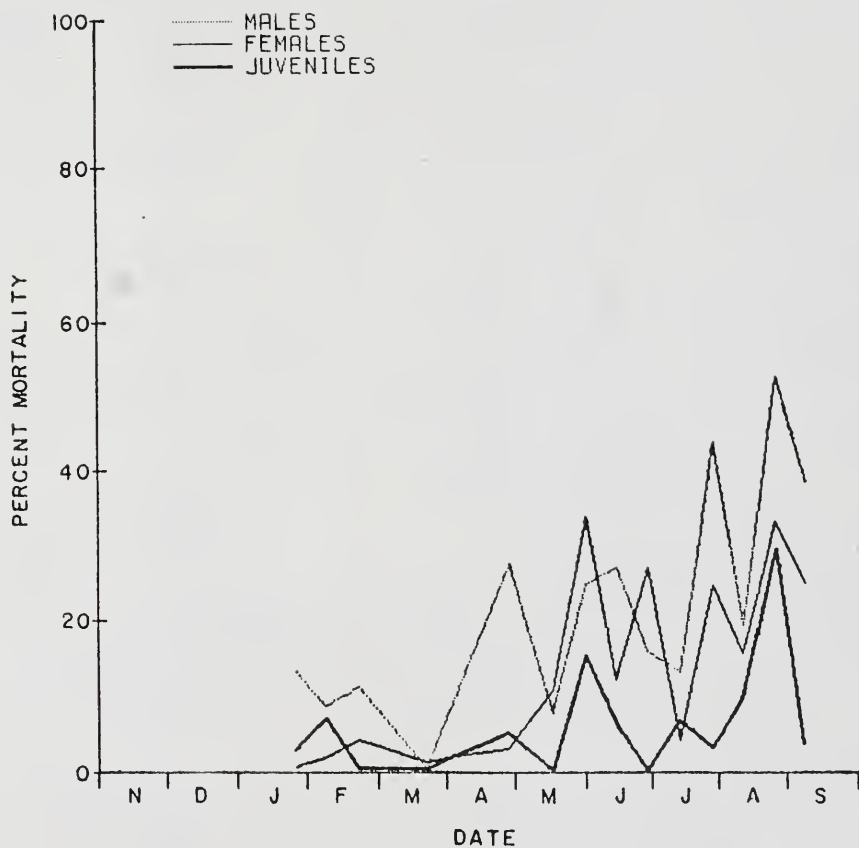


Fig. 6. Seasonal variation in mortality of Acartia tonsa exposed to treatment 100.

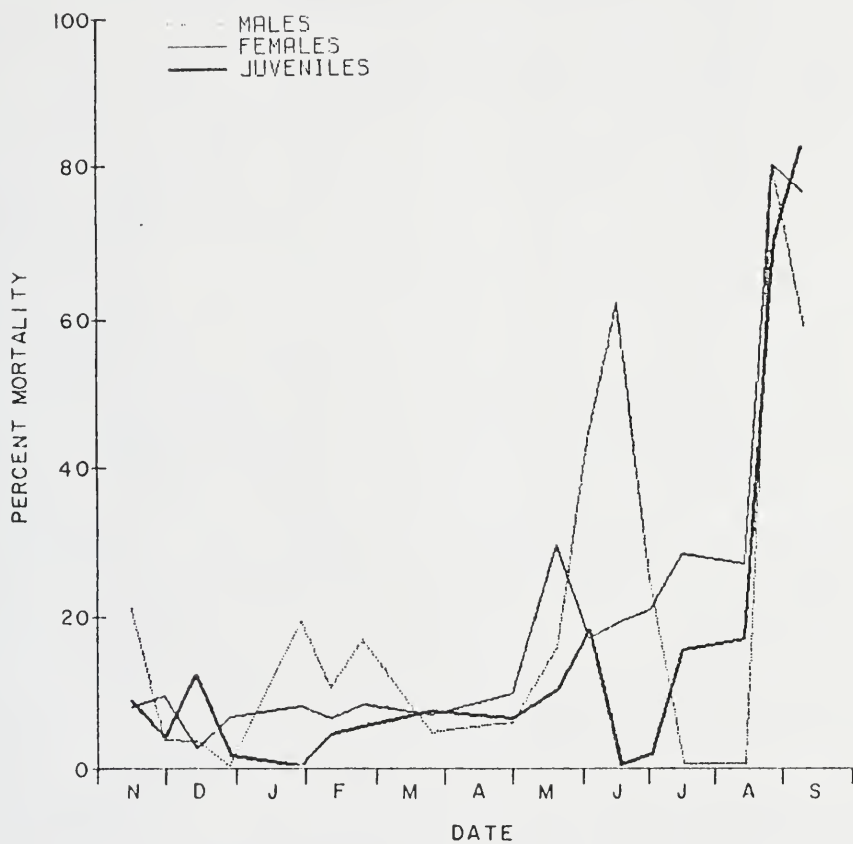


Fig. 7. Seasonal variation in mortality of *Acartia tonsa* exposed to treatment ID2.

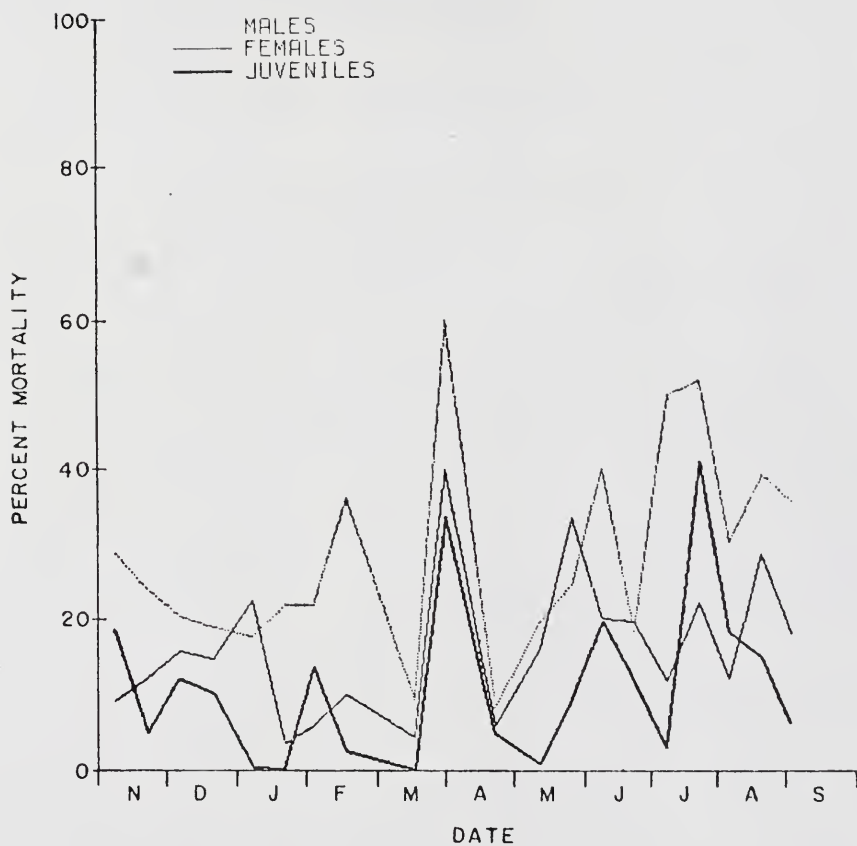


Fig. 8. Seasonal variation in mortality of *Acartia tonsa* exposed to treatment DS0.

rising in mid-summer. Males appear more sensitive than females, with mortalities rising faster than those of the females, while juvenile values lagged behind those of the adults (Figure 9).

Figure 10a shows how the Acartia tonsa mortalities for treatment DS2 are related to discharge temperatures. Mortalities are generally low for discharge temperatures below 35°C and rise with higher temperatures. The increase in mortality appears to be exponential above this temperature.

The predictive regression model chosen by the stepwise process for Acartia tonsa was significant at the .0001 level ($F = 30.87$, 17df; $R^2 = .74$). The response surfaces that were plotted from the regression equation for various conditions are shown in Figure 11.

The response patterns for Acartia tonsa exposed only to thermal shock showed some interesting temperature-salinity interactions. Male A. tonsa (Figure 11a) exhibited higher mortalities than the females (Figure 11b), which in turn were more sensitive than the juveniles (Figure 11c). Similar trends were seen for each category, however, with salinities between 19 and 23 ppt. allowing survival over a broad range of temperature (20 to 35°C). Relatively low mortalities are seen for combinations of high salinities and moderately high temperatures (30 to 35°C). Temperature increases above 35°C, however, are positively correlated with great increases in mortality of the entire range of salinities.

Exposure to the delayed thermal effects caused predicted mortalities to rise slightly in all areas except for conditions of temperature less than 35°C and salinities of 20 to 25 ppt. (Figure 11d). The temperature level at which the exponential rise in mortalities occurs

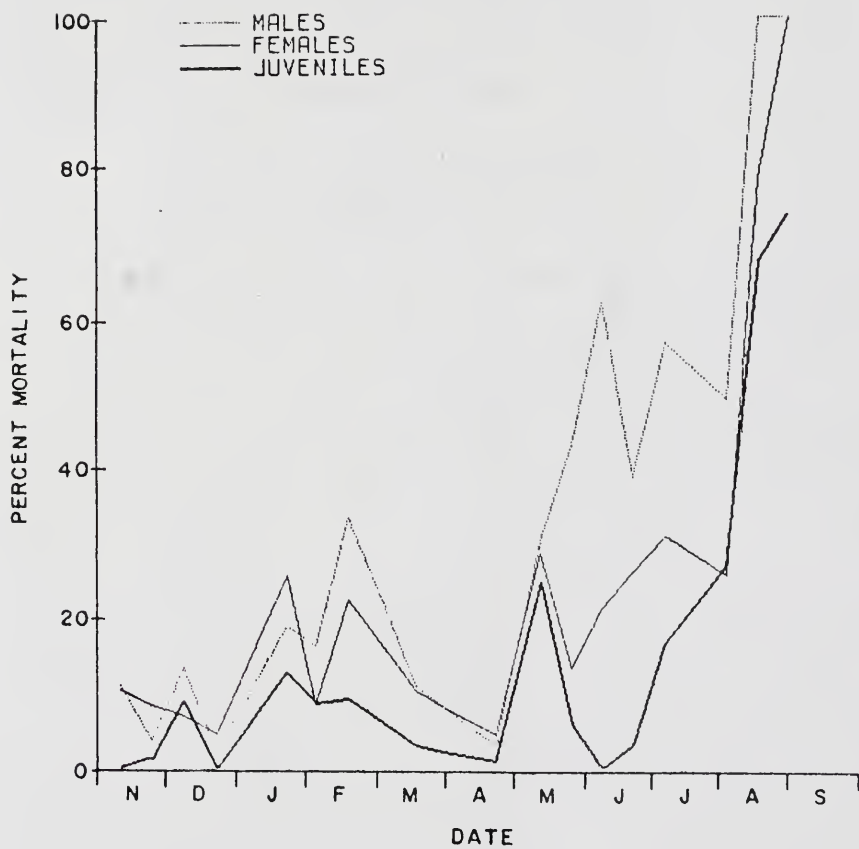


Fig. 9. Seasonal variation in mortality of Acartia tonsa exposed to treatment DS2.

Fig. 10. Entrainment mortality as a function of discharge temperature: (a) Acartia tonsa; (b) Oithona spp.; (c) Paracalanus crassirostris; (d) Euterpina acutifrons; (e) Labidocera spp.; (f) Pseudodiaptomus coronatus.

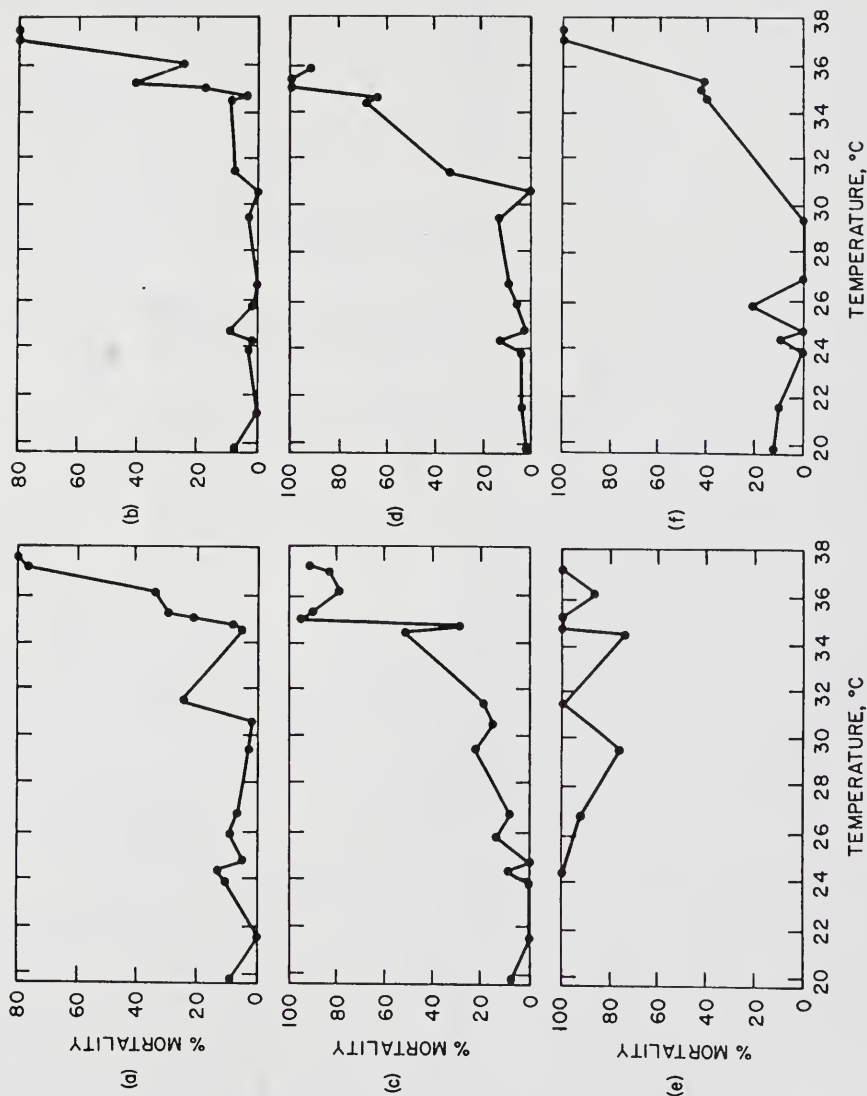


Fig. 10. Continued
(g) Tortanus setacaudatus; (h) total
copepod population.

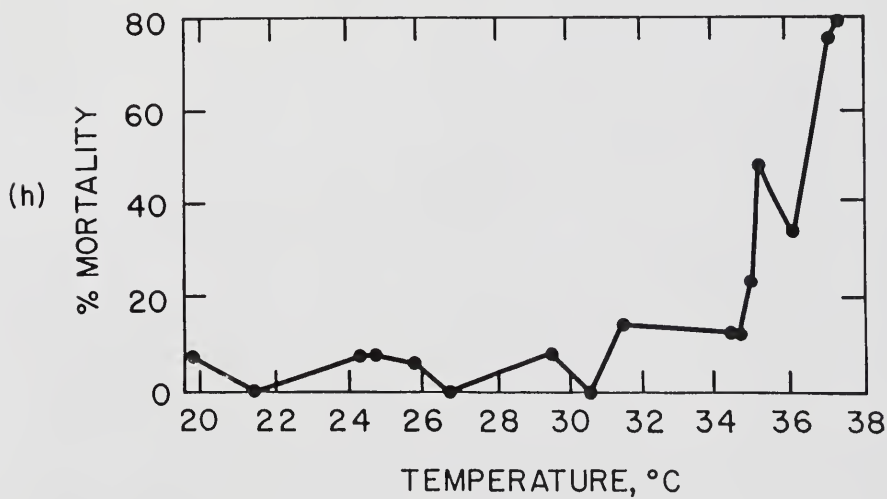
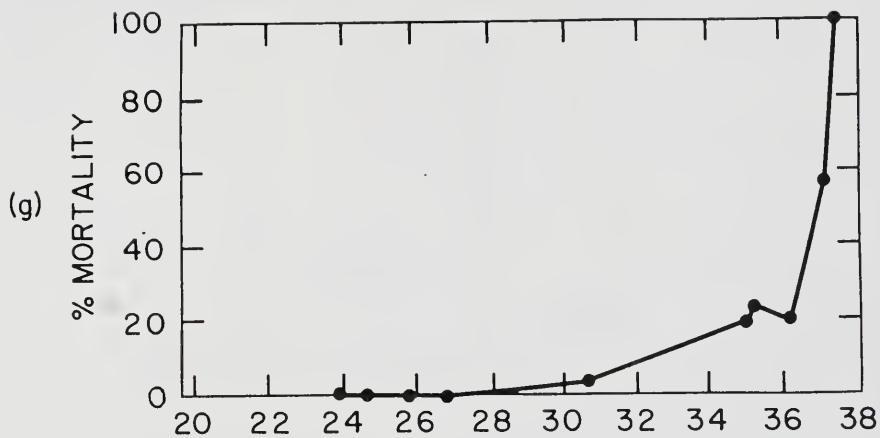
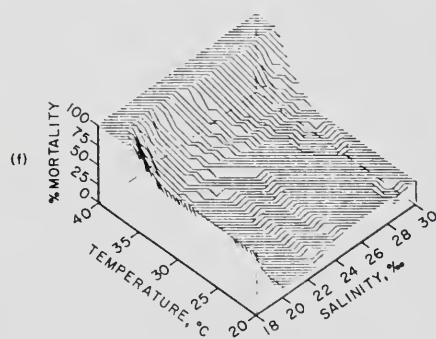
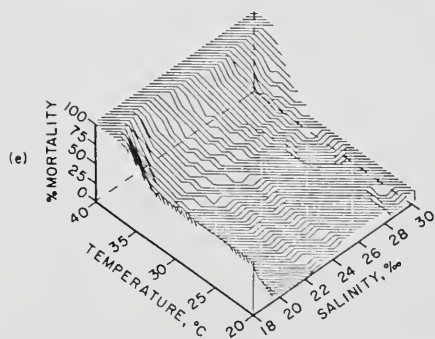
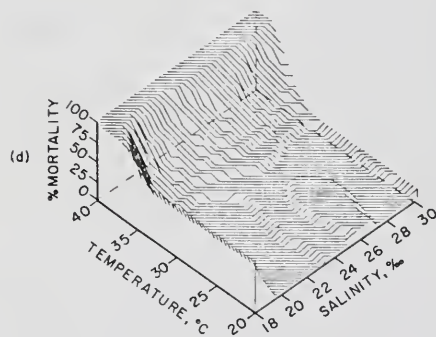
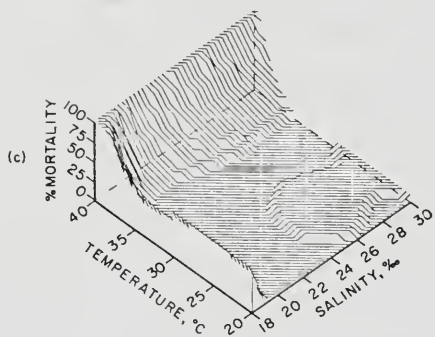
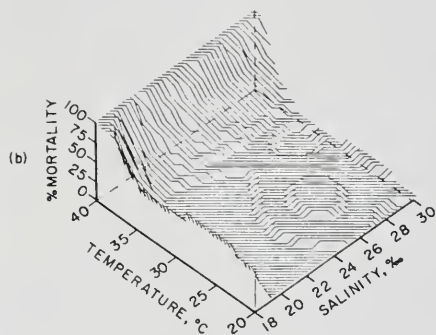
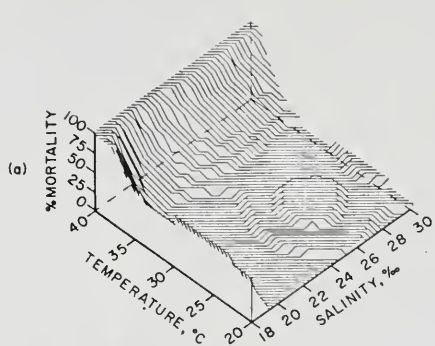


Fig. 11. Response surface estimates of mortality as a function of discharge temperature and salinity for populations of Acartia tonsa exposed to various conditions: (a) male, (b) female and (c) juvenile immediate response to thermal shock; (d) female response to extended exposure to elevated temperatures; (e) female response to mechanical damage in addition to delayed thermal effects; and (f) female response to the same conditions in spring.



is lowered below 35°C in areas of higher salinities and the range of temperatures at which 100% mortality occurs has been broadened.

When mechanical effects are considered into the entrainment conditions (Figure 11e), a rise in mortality of 3 to 5% occurs in all areas except those of optimum conditions (20 to 35°C , 20 to 23 ppt.). The Acartia tonsa male response pattern (not shown) exhibits a similar trend except increases in predicted mortality are nearer 10%.

The addition of seasonal effects associated with spring produces a response pattern for Acartia tonsa (Figure 11f) that looks identical to those for other seasons (Figure 11e), except that all predicted mortality values are approximately 10% higher.

Oithona spp.

Oithona spp. is a category made up of at least three species of the genus Oithona: O. brevicornis, O. nana and O. simplex. Species identification was not feasible for the males and juveniles of this genus under the magnifications used for counting. Positive species identification involves dissection of the mouth parts and observation under a compound microscope (Grice, 1960a). Oithona brevicornis was, however, by far the most dominant of these species whenever identifications were made. Therefore, it is assumed that this species makes up a majority of the category throughout the year.

Oithona spp. are the numerically dominant species of copepod in the Crystal River area. Females of these species ranged in numbers from $492/\text{m}^3$ to $17,819/\text{m}^3$ with a mean of $5,298/\text{m}^3$ (Figure 12). The males were much rarer, with numbers from $61/\text{m}^3$ to $1,151/\text{m}^3$ and a mean

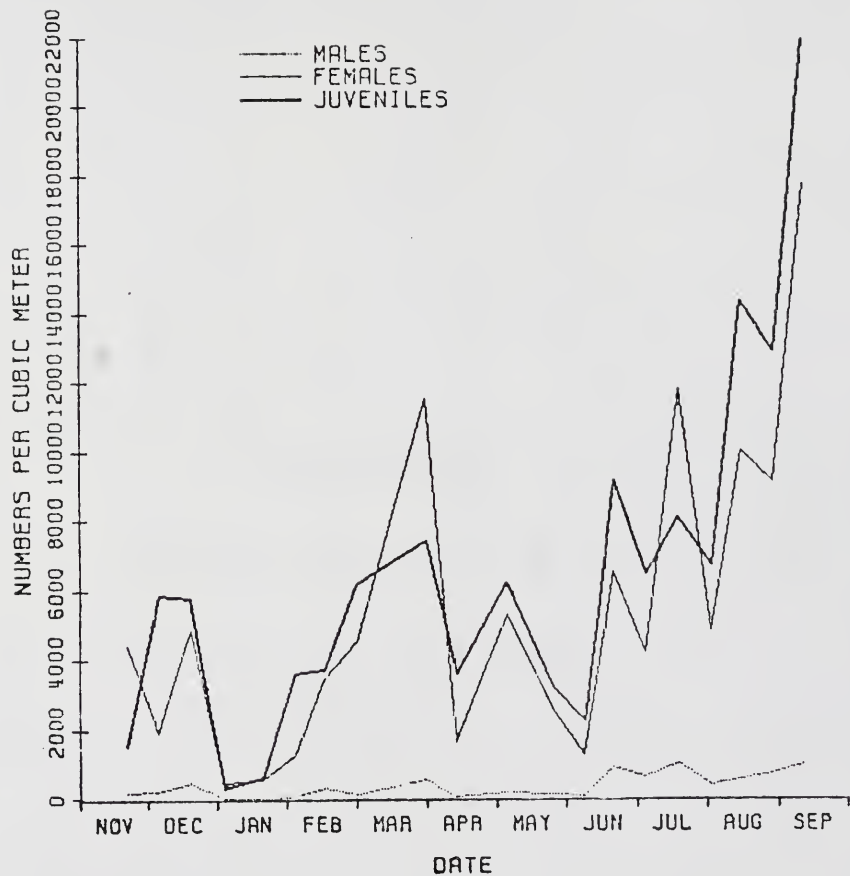


Fig. 12. Seasonal variation in numbers of *Oithona* spp.

of $477/\text{m}^3$. Juveniles ranged in numbers from $341/\text{m}^3$ to $22,067/\text{m}^3$, with a mean of $7,136/\text{m}^3$. Peak numbers occurred in December, March and September, with a fairly steady increase in numbers from June to the time of highest densities at the end of the summer. Oithona spp., as a whole, represents from 20% to nearly 80% of the total population of copepods (Figure 13). Females and juveniles of these species nearly always accounted for at least 50% of the numbers of the total copepod population.

Mortalities for Oithona spp. which had undergone the thermal shock of the ID0 treatment were from 0 to 52%, with a year-round mean of 6.55%. On the dates that showed peak mortalities, male mortality values were higher than those of females or juveniles (Figure 14).

The Oithona spp. populations that had traveled down the discharge canal following the initial thermal shock (treatment ID2) exhibited an average mortality of 13.4% with a range from 0 to 74%. Mortality values remained relatively low until mid-summer when they rose rapidly to the highest levels (Figure 15). Again, males showed higher mortalities than females or juveniles, rising to the 100% level by mid August.

Populations of Oithona spp. sampled immediately upon leaving the power plant (treatment DS0) had mortalities from 0 to 44% with a mean of 8.6%. Mortality values fluctuated around a relatively low level until mid-summer, when they rose (Figure 16). Differential mortality due to sex or age class was only observed during these warmer months when male mortalities were greater.

Oithona spp. that were exposed to the full effects of entrainment through treatment DS2 had a range of mortalities from 0 to 80% and a mean of 16%. Mortalities remained low until April, after which time

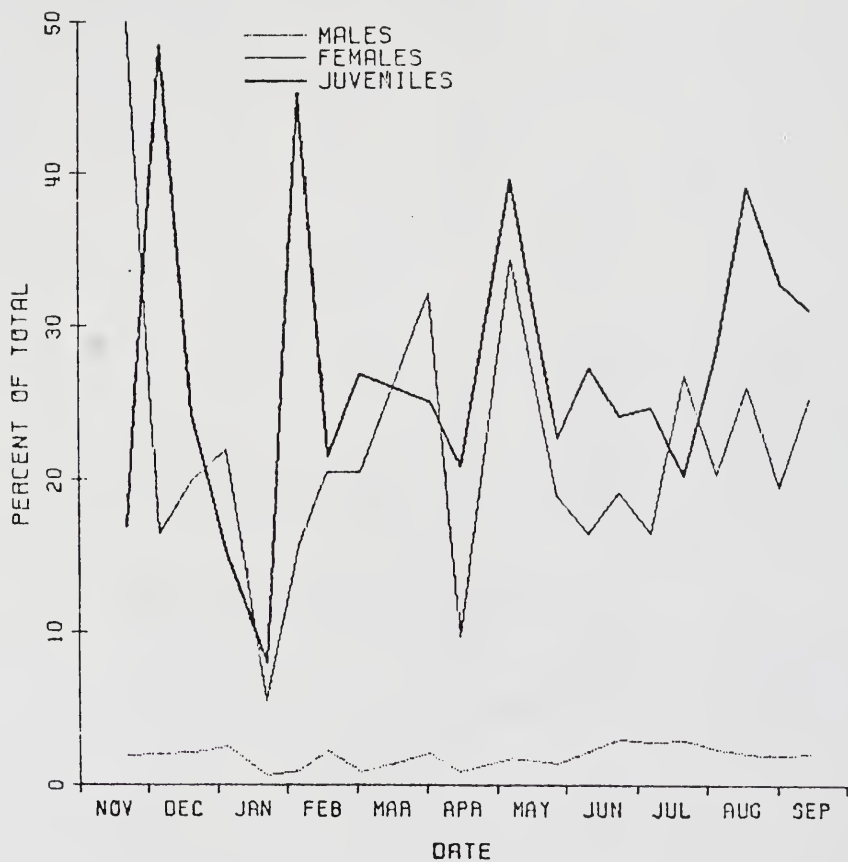


Fig. 13. Seasonal variation in percent of total copepod population represented by *Oithona* spp.

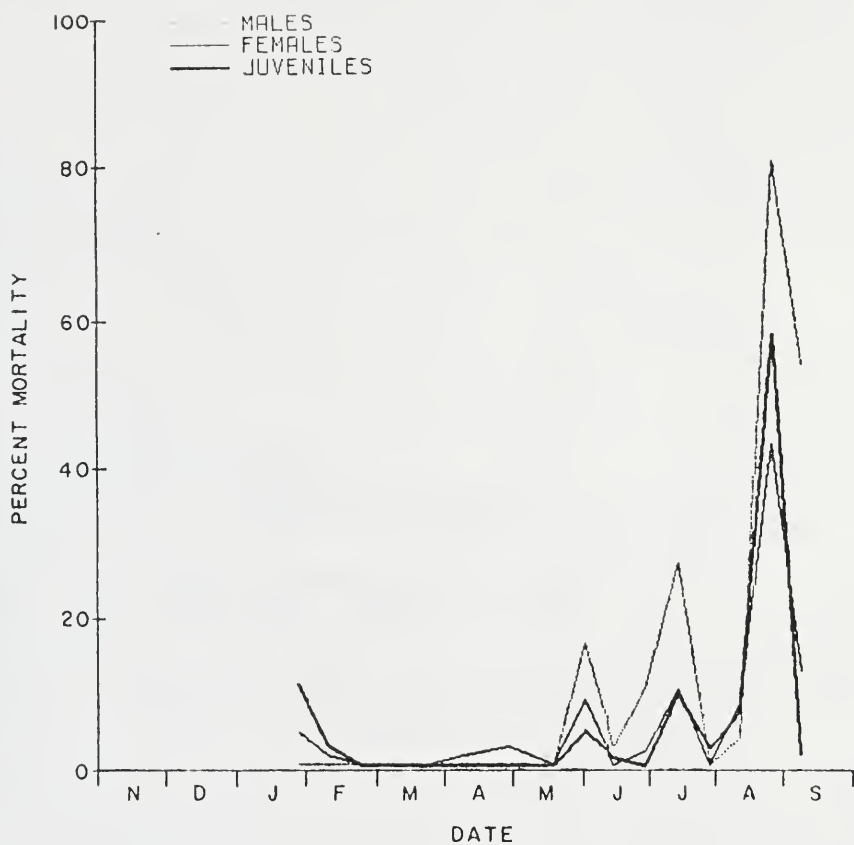


Fig. 14. Seasonal variation in mortality of *Oithona* spp. exposed to treatment ID0.

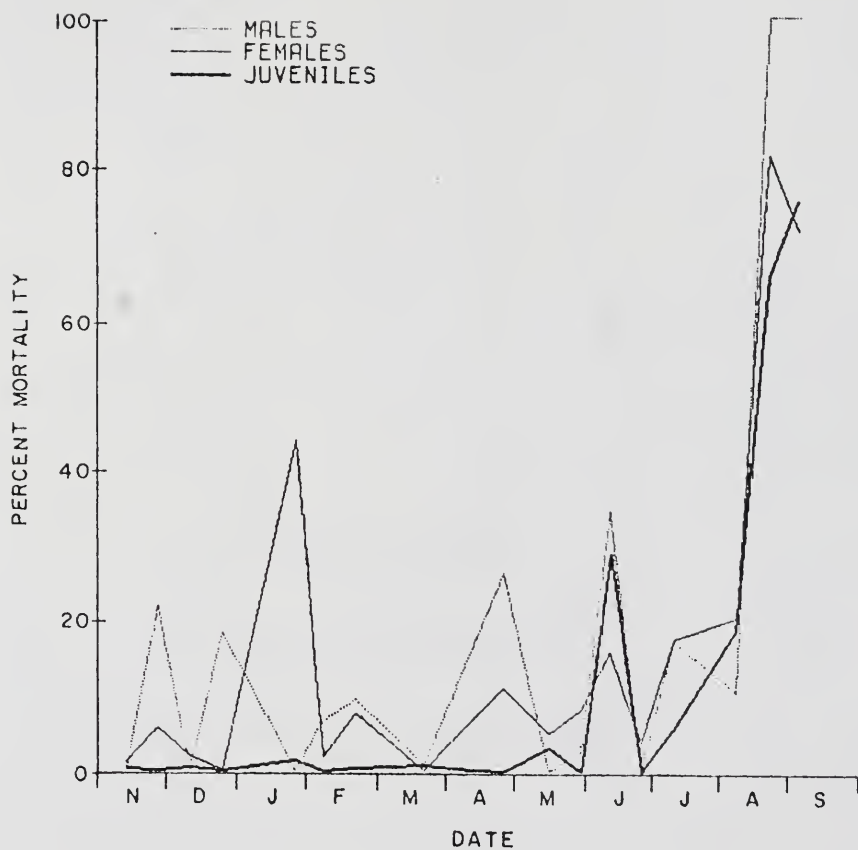


Fig. 15. Seasonal variation in mortality of *Oithona* spp. exposed to treatment 1D2.

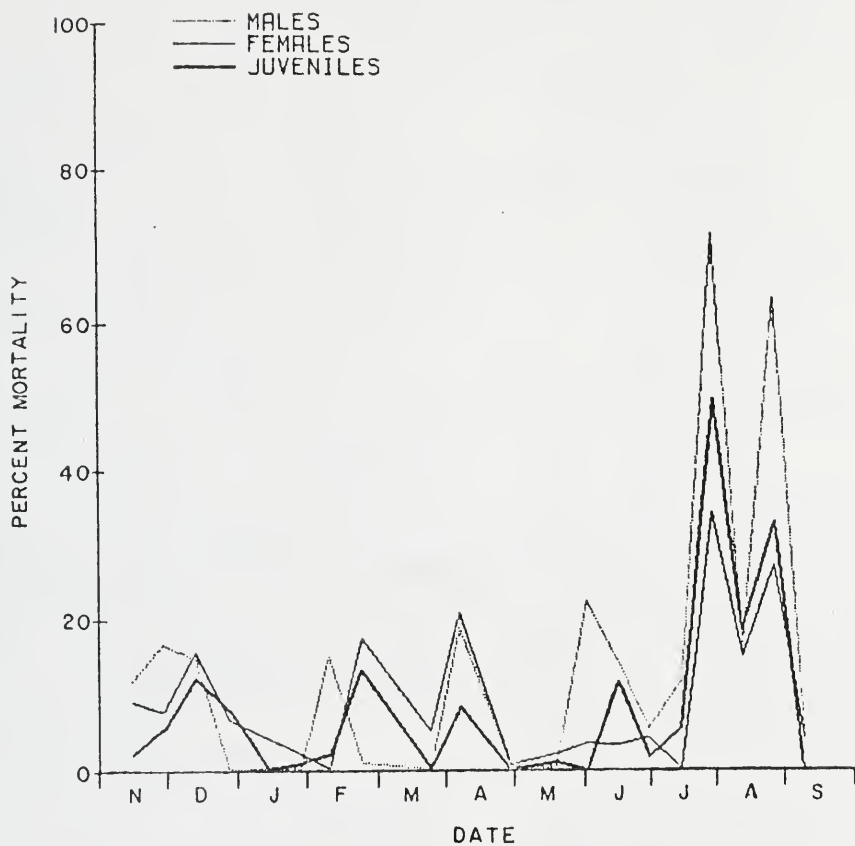


Fig. 16. Seasonal variation in mortality of *Oithona* spp. exposed to treatment DS0.

there was a gradual rise through the month of June (Figure 17). In July there was a sharp increase in mortality, ending with the highest values by the end of summer. Males showed greater sensitivity to mortality than females or juveniles when exposed to this treatment.

The relationship of the entrainment mortality of Oithona spp. to discharge temperatures is shown in Figure 10b. Consistently low mortality values were seen until the discharge temperatures rose above 35°C. Above the 35°C level, mortalities increased rather rapidly with rising temperatures.

The predictive regression model for Oithona spp. was significant at the .0001 level ($F = 44.25$, 12df; $R^2 = .74$). The response surfaces that were plotted from the regression equation for various conditions are shown in Figure 18.

Female and juvenile Oithona (Figure 18a) show low (0 to 20%) mortalities when exposed to thermal shock if the discharge temperatures are below 35°C. Higher salinities apparently buffer the effects of moderately high temperatures (20 to 35°C). Above 35°C, however, mortalities rise exponentially with small increases in temperature, regardless of salinities. Male Oithona (Figure 18b) exhibit similar patterns, but appear to be more sensitive to the thermal shock than females or juveniles, having slightly higher mortality values for all conditions except for the very lowest temperatures (20 to 22°C).

The extended exposure (2 hours) to the heated effluents in the discharge canal causes the male (Figure 18c) and female-juvenile (Figure 18d) response patterns to change. Mortalities rise for conditions of higher temperature (above 32°C), especially with low salinities, since the high salinity-high temperature resistance synergism is

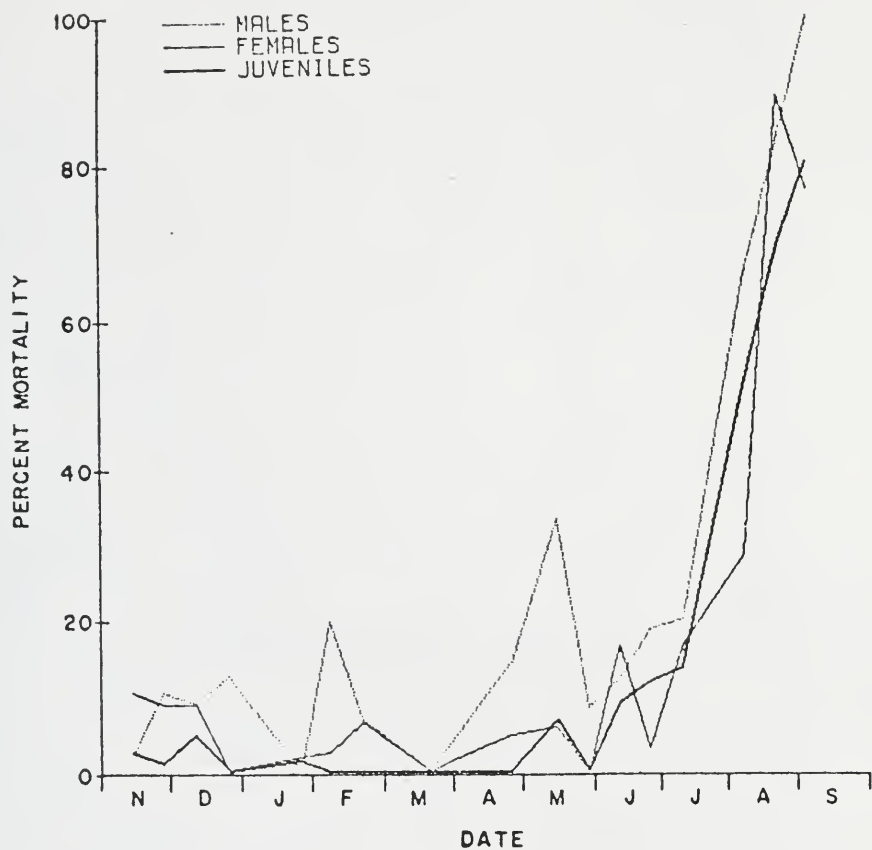
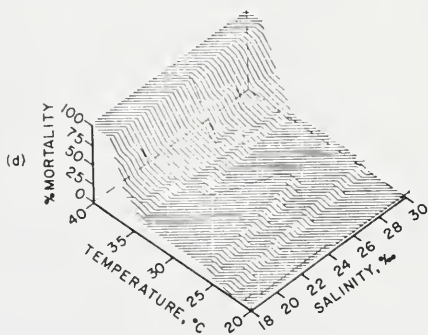
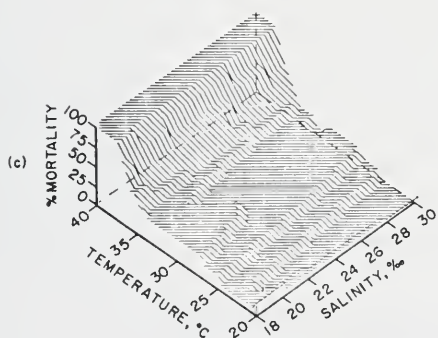
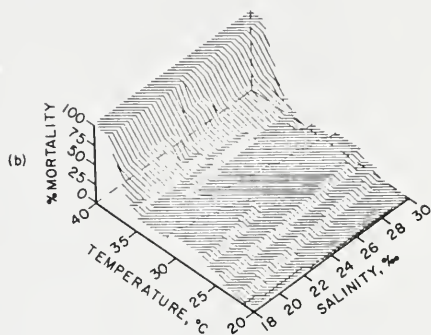
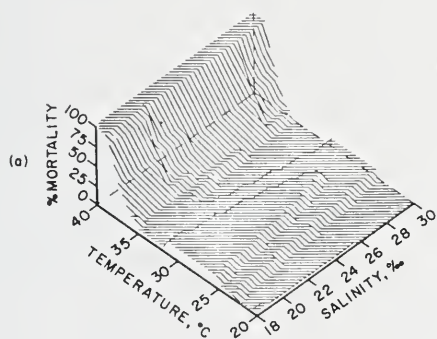


Fig. 17. Seasonal variation in mortality of *Oithona* spp. exposed to treatment DS2.

Fig. 18. Response surface estimates of mortality as a function of discharge temperature and salinity for populations of *Oithona* spp. exposed to various conditions: (a) female and (b) male immediate response to thermal shock; (c) male and (d) female response to extended exposure to elevated temperatures.



still somewhat evident. The temperature where 100% mortality occurs has also been lowered because of these delayed thermal effects within the discharge canal.

Mechanical, seasonal and density dependent factors were shown to be insignificant in relation to entrainment mortality for Oithona spp.

Paracalanus crassirostris

Paracalanus crassirostris is a small calanoid copepod that was the third most abundant copepod in the Crystal River area. Males of this species ranged from $13/\text{m}^3$ to $1,037/\text{m}^3$ with a mean value of $422/\text{m}^3$ for the year (Figure 19). Females were generally more abundant, with a range of from $40/\text{m}^3$ to $2,793/\text{m}^3$ and a mean of $848/\text{m}^3$. Juvenile P. crassirostris ranged in numbers from $35/\text{m}^3$ to $4,382/\text{m}^3$ and had a mean value of $2,583/\text{m}^3$. Peak numbers of this species occurred in March, May, June and September. Paracalanus crassirostris represents between 5 to 48% of the copepods of the area throughout the year and generally accounts for 10 to 20% of the total population (Figure 20).

The population of Paracalanus crassirostris exposed to treatment ID0 had mortality values from 0 to 78% and a mean value of 18.2%. Mortalities were low from January through May, then increasingly rose to high levels by mid August (Figure 21). No trends in differential mortality attributable to sex or age class were observed for this treatment.

Paracalanus crassirostris that were left in the heated effluent for several hours following the initial shock (treatment ID2) showed higher mortality values, with a range from 0 to 87% and a mean of 22.7%.

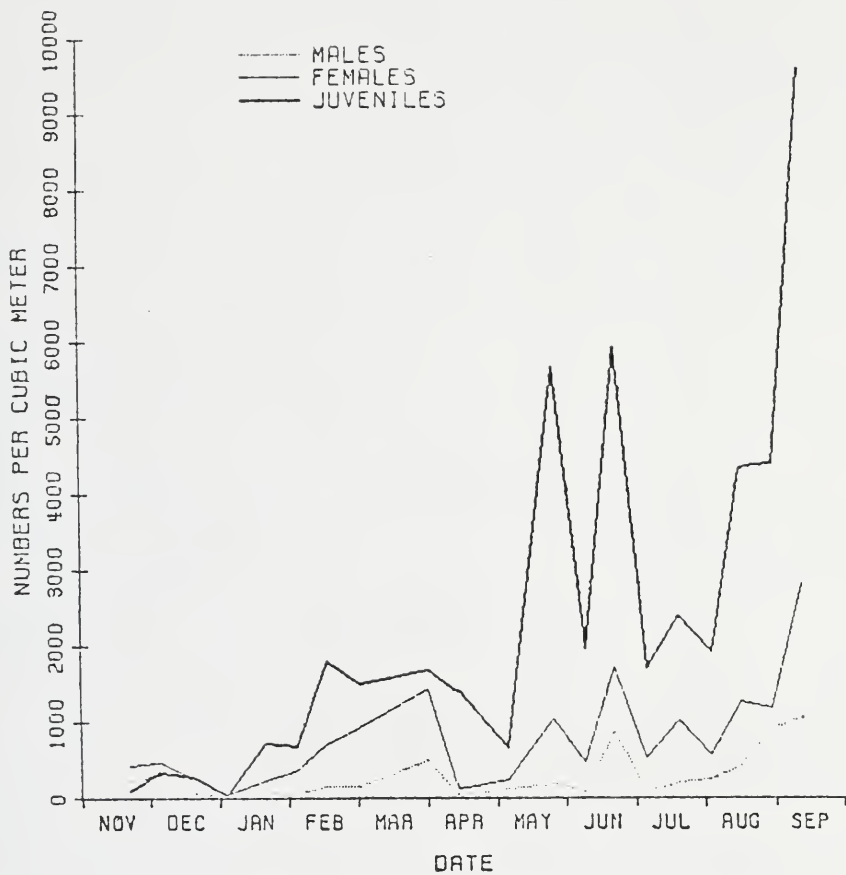


Fig. 19. Seasonal variation in numbers of Paracalanus crassirostris.

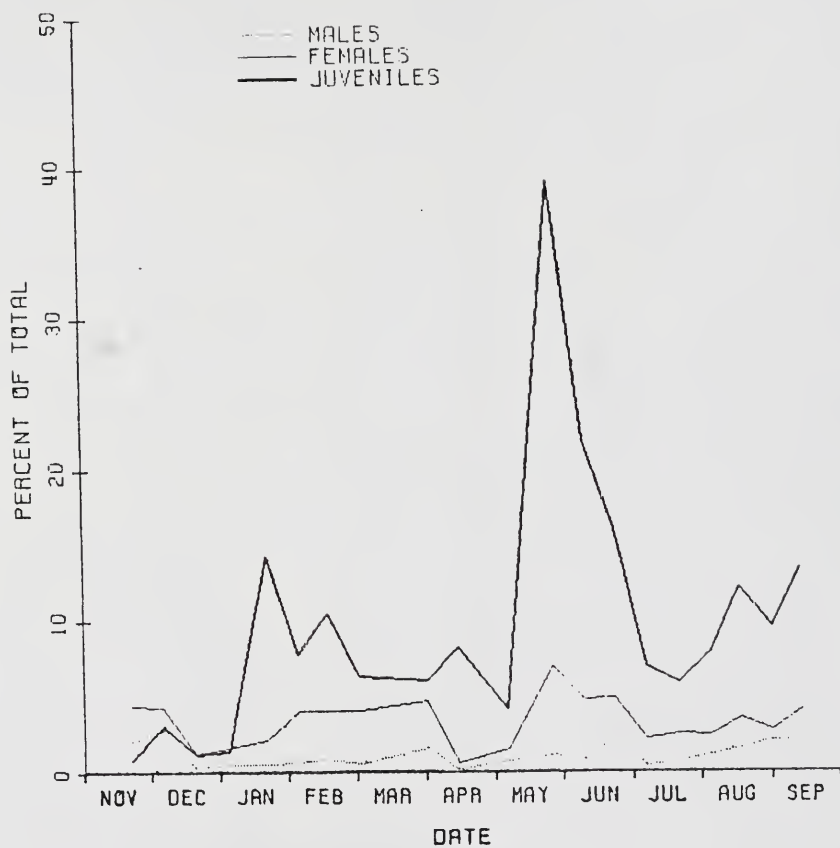


Fig. 20. Seasonal variation in percent of total copepod population represented by *Paracalanus crassirostris*.

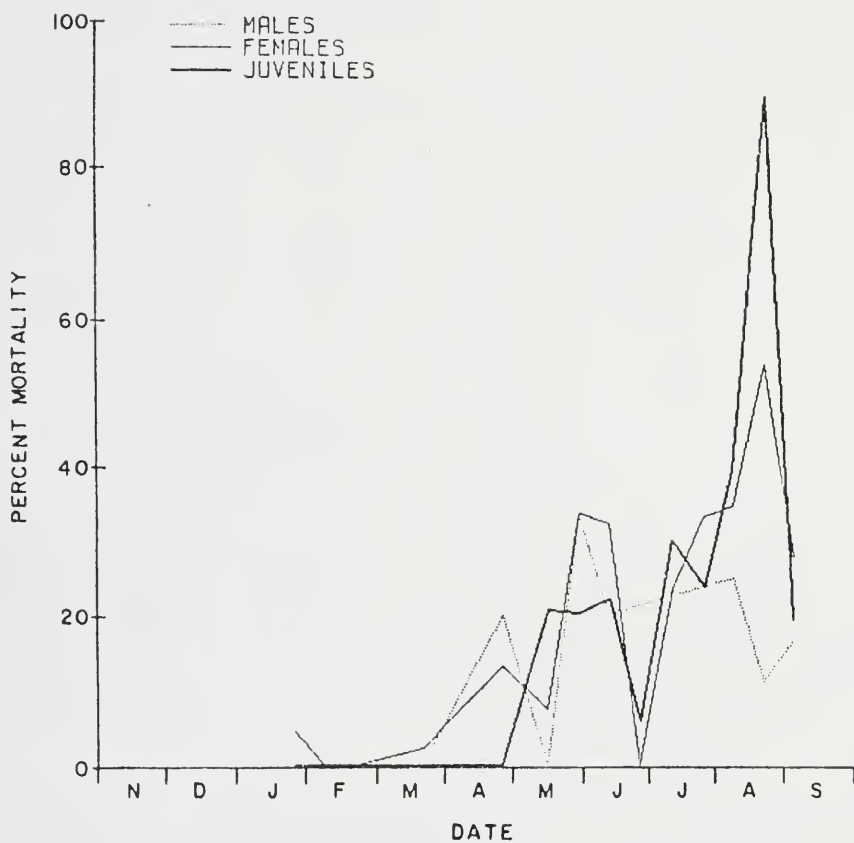


Fig. 21. Seasonal variation in mortality of *Paracalanus crassirostris* exposed to treatment 1D0.

The same general trend in mortality was seen for this treatment, with low mortalities through the winter and spring seasons, followed by a rise in early summer and a very rapid rise by mid-summer (Figure 22). With this treatment, males tended to have higher mortalities during the warmer months than females or juveniles.

Immediate bioassay of Paracalanus crassirostris leaving the power plant (treatment DS0) showed an annual mean mortality of 24.7% and a range of values from 1 to 83%. Mortalities fluctuated within the 15 to 20% range for most of the winter and spring, then began to rise after mid April (Figure 23). A peak in mortalities was observed in July.

The populations of Paracalanus crassirostris subjected to two hours in the heated waters of the discharge canal following entrainment (treatment DS2) had a mean mortality of 31% and a range of 0 to 100%. Mortalities fluctuated at a low level through March, then rose at an increasingly rapid rate to highest values by early summer (Figure 24). Males appeared to be slightly more sensitive than females or juveniles during this period of rising mortalities.

Entrainment mortality of Paracalanus crassirostris is shown in relationship to discharge temperatures in Figure 10c. Mortalities increased gradually up to 31°C and then exhibited a progressively steeper rise to high levels with higher temperatures.

The predictive regression model for Paracalanus crassirostris was significant at the .0001 level ($F = 38.97, 11df; R^2 = .76$). The response surfaces plotted from the regression equation for various conditions are shown in Figure 25.

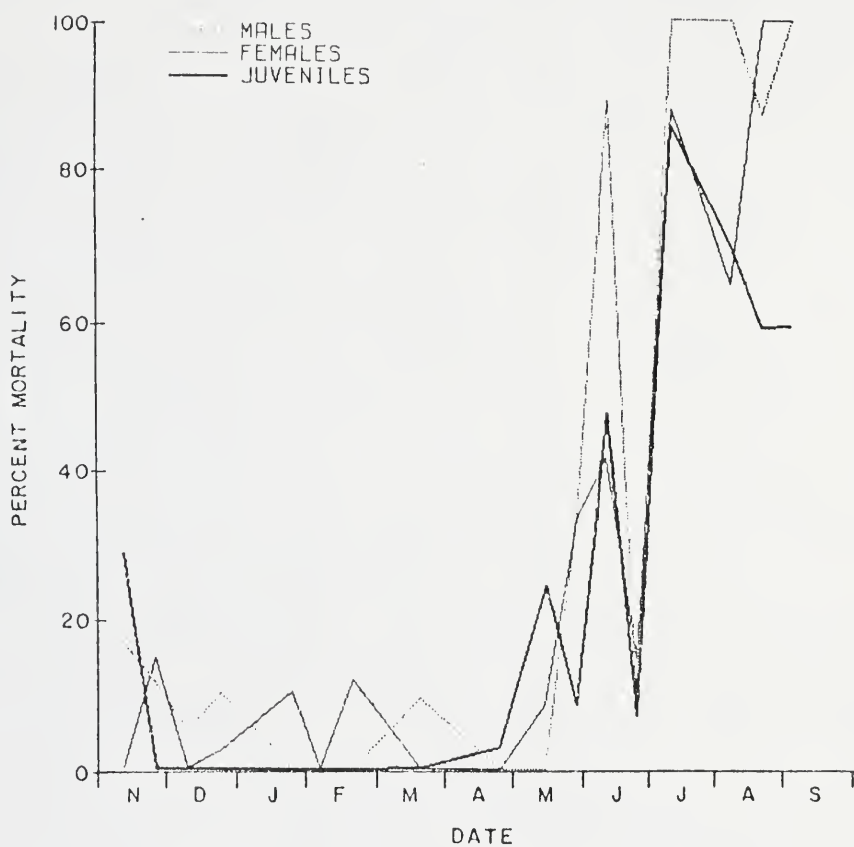


Fig. 22. Seasonal variation in mortality of Paracalanus crassirostris exposed to treatment ID2.

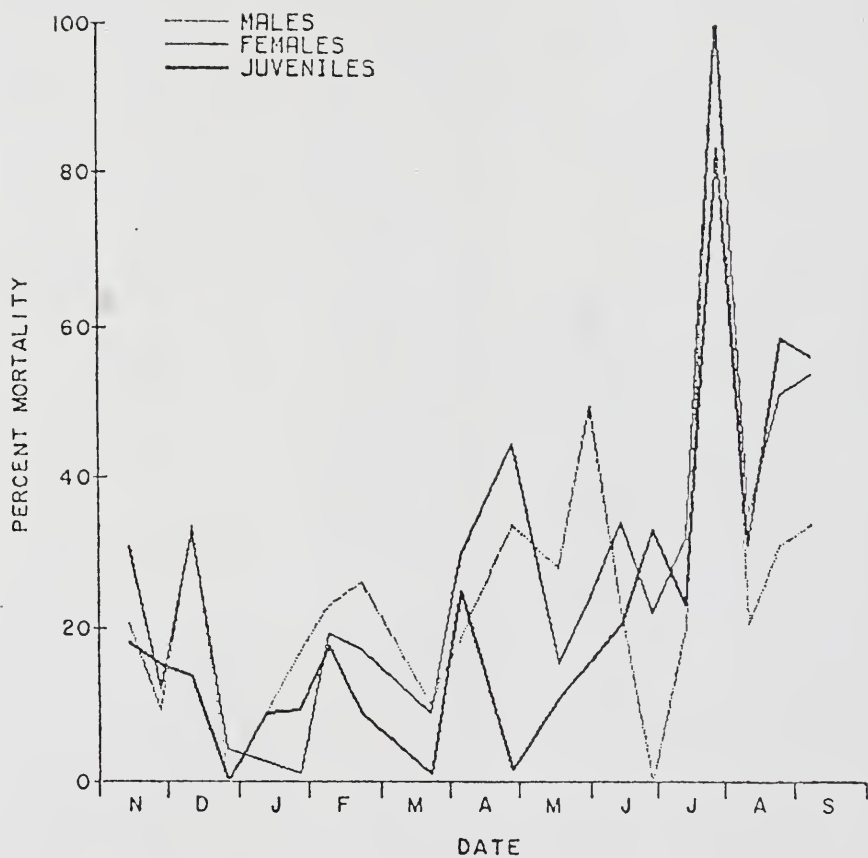


Fig. 23. Seasonal variation in mortality of *Paracalanus crassirostris* exposed to treatment DSO.

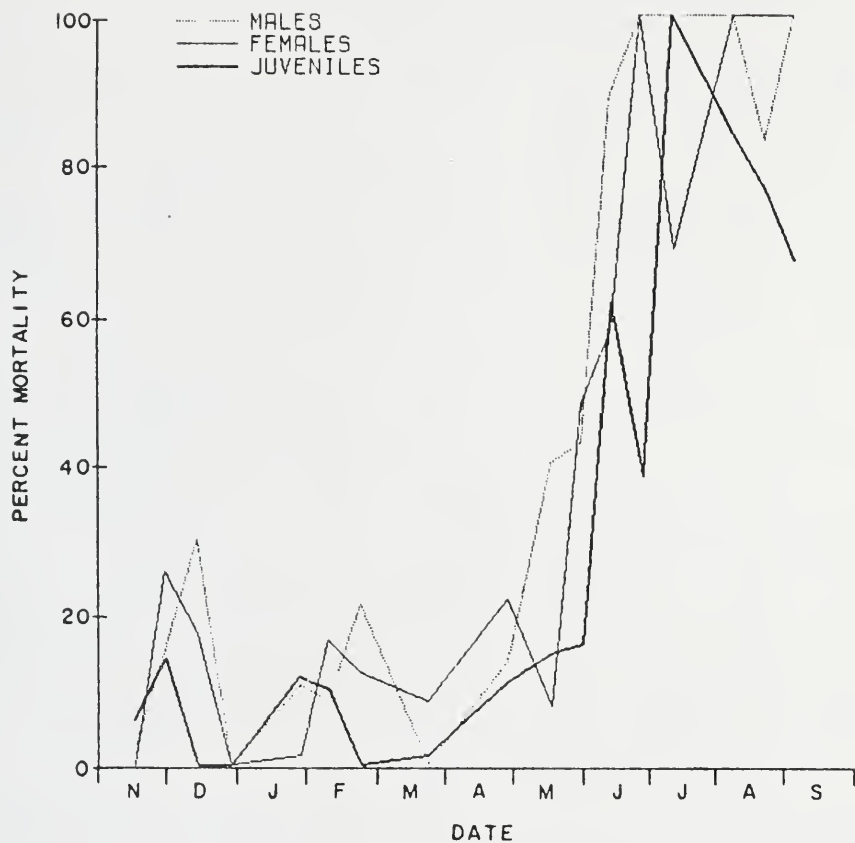
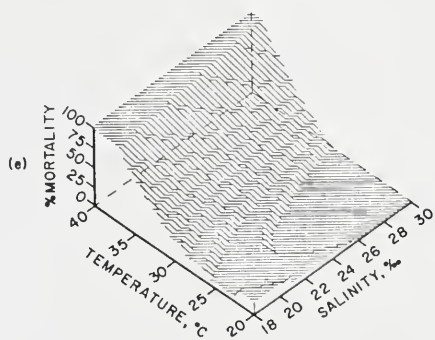
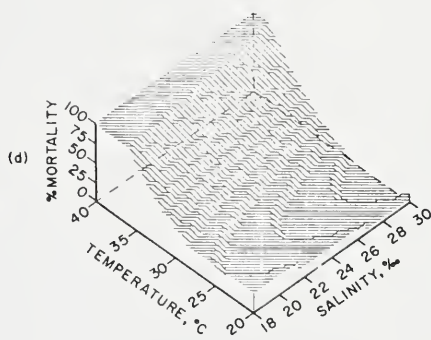
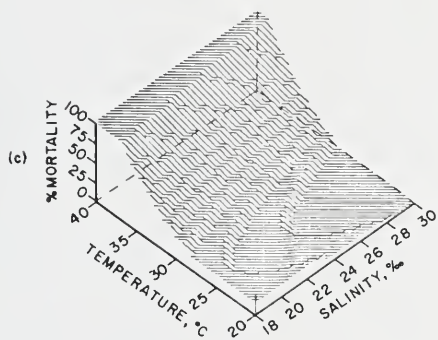
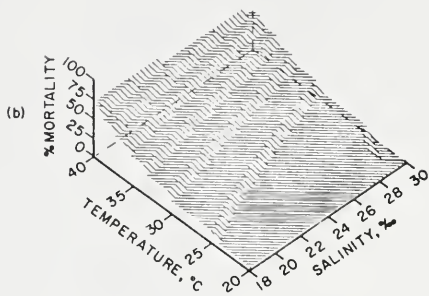
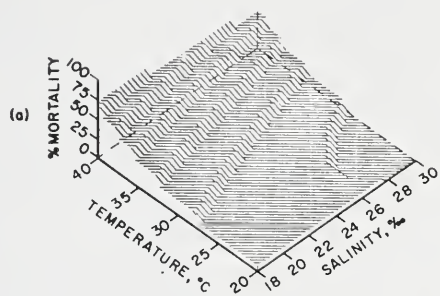


Fig. 24. Seasonal variation in mortality of Paracalanus crassirostris exposed to treatment DS2.

Fig. 25. Response surface estimates of mortality as a function of discharge temperature and salinity for populations of Paracalanus crassirostris exposed to various conditions: (a) male and (b) female-juvenile immediate response to thermal shock; (c) female response to extended exposure to elevated temperatures; (d) female response to mechanical damage in addition to delayed thermal effects; and (e) female response to the same conditions in spring.



Paracalanus crassirostris males (Figure 25a) showed slightly lower mortalities than did the females and juveniles (Figure 25b) when exposed to thermal shock. The response patterns show a gradually increasing rise in mortalities with increases in temperature. A slight temperature-salinity interaction is evident throughout the range of conditions with higher salinities producing lower mortalities for any temperature. Mortality values for P. crassirostris do not rise much above 70%, even for highest temperatures (40°C) during this short-term exposure to thermal stress.

Extended exposure to the heated effluent (Figure 25c) causes Paracalanus crassirostris mortality values to rise rapidly for temperatures above 30°C . For temperatures below this point, higher salinities continue to produce lower mortalities. Mortalities of the males subjected to extended exposure were 10 to 15% higher than those of females and juveniles.

When the mechanical effects of entrainment are included in the Paracalanus crassirostris response surface equation (Figure 25d), mortalities are only slightly increased for temperatures below 30°C . Above this point, the rise to the 100% mortality level occurs more rapidly and for lower temperature values than would be seen for a population not subjected to the physical factors involved in passage through the plant (Figure 25c).

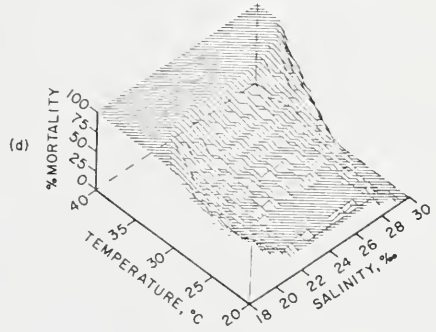
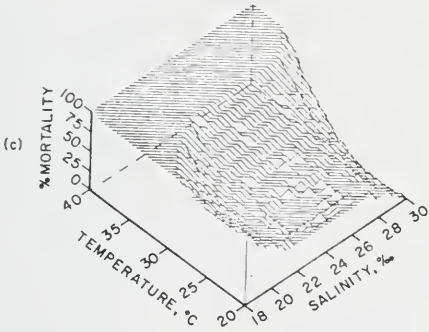
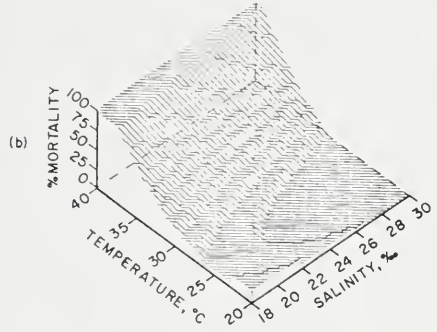
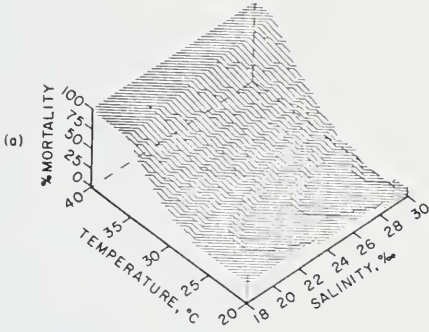
The Paracalanus crassirostris populations observed during the spring (Figure 25e) had mortalities approximately 10% lower than would be expected for all conditions, except the most extreme temperatures (above 37°C).

Paracalanus crassirostris response patterns are significantly affected by the density (numbers/m³) of the species at the time of entrainment. When numbers per cubic meter are low (Figure 26a), entrainment mortalities are generally higher than when the numbers are high (Figure 26b). This effect is particularly evident for the more lethal conditions, where the temperature at which the 100% mortality level occurs may be changed several degrees by the density factor.

Euterpina acutifrons

Euterpina acutifrons is a pelagic harpacticoid that was the fourth most abundant species in the Crystal River area. Females ranged in numbers from 5/m³ to 857/m³ and had a mean of 177/m³ (Figure 27). Male E. acutifrons were rarer, ranging in abundance from 0/m³ to 184/m³ and having a mean value of 60/m³. Numbers for juveniles of this species ranged from 0/m³ to 2,617/m³, with a mean of 944/m³. There were three obvious peak periods for this species: one in December, a second at the end of February, and the third at the beginning of June. By mid-summer months, however, E. acutifrons had become scarce in the samples. The trend toward the decreasing importance of this species with the warmer months is reflected in the percent of the total population that it represented through the year (Figure 28). Through most of the winter and spring months this species accounted for 5 to 10% of the copepods of the area but gradually decreased in importance until, by mid-summer, it represented only a fraction of a percent of the total population.

Fig. 26. Response surface estimates of mortality as a function of discharge temperature and salinity for entrainment populations at different densities: (a) female Paracalanus crassirostris at lowest numbers per m³; (b) female Paracalanus crassirostris at highest numbers per m³; (c) female Euterpina acutifrons at lowest numbers per m³; (d) female Euterpina acutifrons at highest numbers per m³.



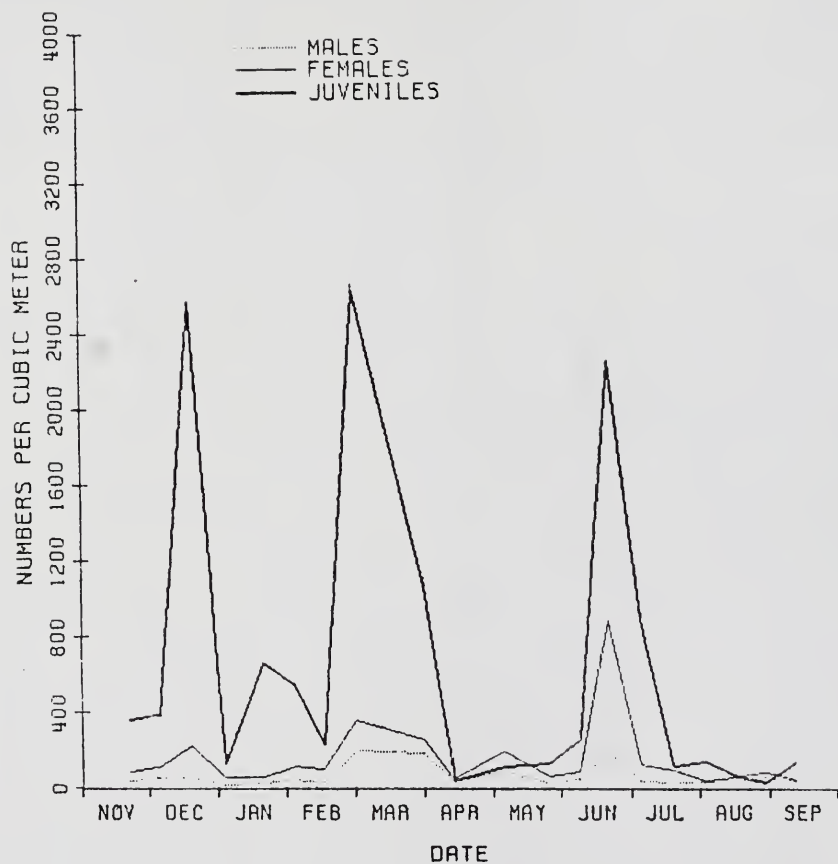


Fig. 27. Seasonal variation in numbers of *Euterpina acutifrons*.

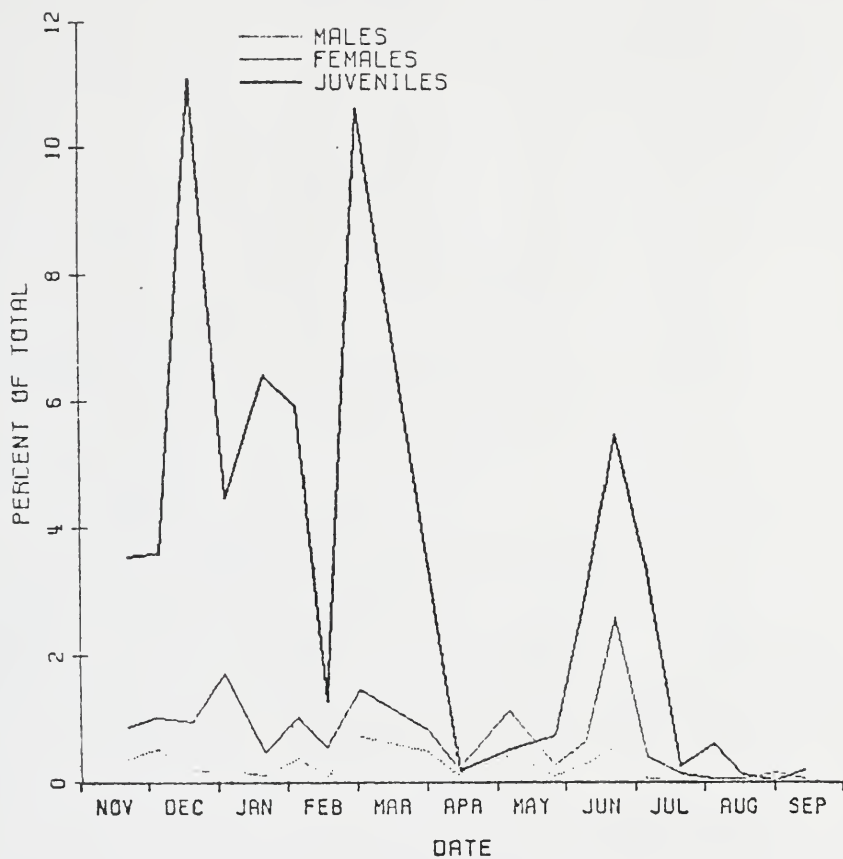


Fig. 28. Seasonal variation in percent of total copepod population represented by Euterpina acutifrons.

Euterpina acutifrons exposed to treatment ID0 exhibited mortality values from 0 to 100% with a mean of 27.7%. Mortalities were generally low until May, when a peak in mortalities occurs (Figure 29). Values then rose again in mid-summer, though scarcity in the numbers of this species after this time prevented further calculations of mortalities.

The mortalities of Euterpina acutifrons subjected to prolonged exposure to the heated effluent (treatment ID2) ranged from 0 to 100% with a mean value of 25%. Mortalities were generally low until mid-spring when they rose steeply to highest levels (Figure 30). Females appeared more sensitive than juveniles to the warmer temperatures, with female mortalities rising to high levels a month earlier in the spring. The trend in male mortality could not be discerned because dwindling numbers with the coming of spring provided only a few data points.

Euterpina acutifrons sampled for bioassay immediately upon leaving the power plant (treatment DS0) showed mortalities that ranged from 0 to 100%, with a mean value of 20%. Mortalities fluctuated at a low level until May when they rose to higher levels (Figure 31). Values rose to the 100% level by the beginning of July, after which no further calculations could be made due to low numbers.

Euterpina acutifrons populations left in the heated waters of the discharge canal following entrainment (treatment DS2) showed a mean mortality value of 33% and a range from 0 to 100%. Mortalities remained quite low through April, but then steeply rose to the 100% level in July (Figure 32). Males showed the increase in mortalities earliest in the spring, followed by the females and then the juveniles.

The relationship of Euterpina acutifrons entrainment mortality to discharge temperatures is shown in Figure 10d. Mortalities were low

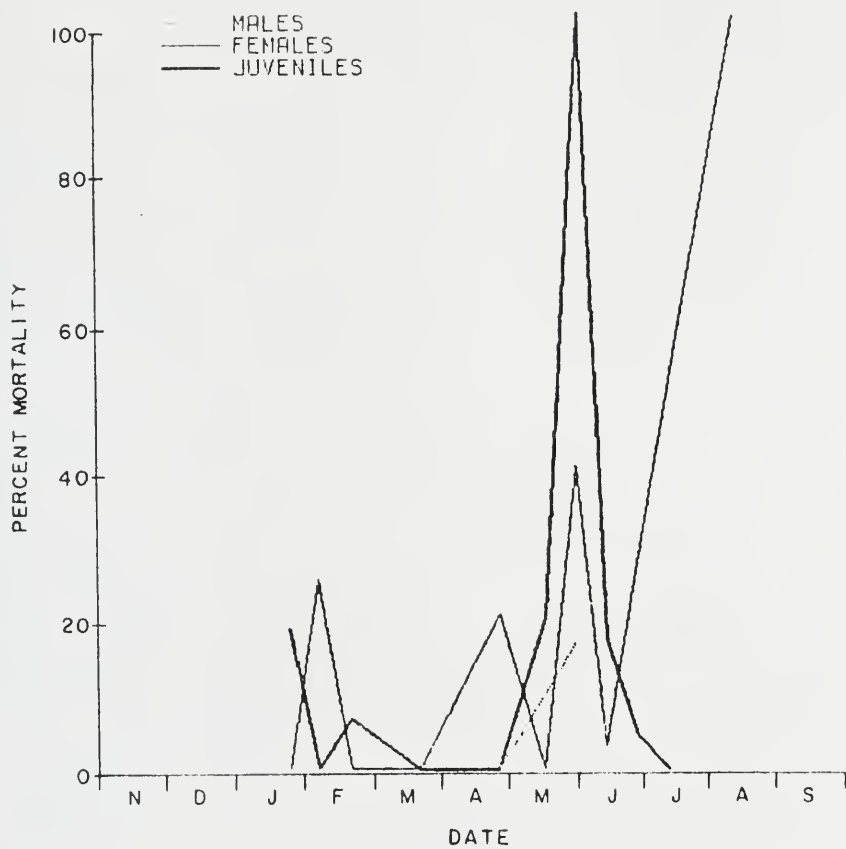


Fig. 29. Seasonal variation in mortality of Euterpina acutifrons exposed to treatment ID0.

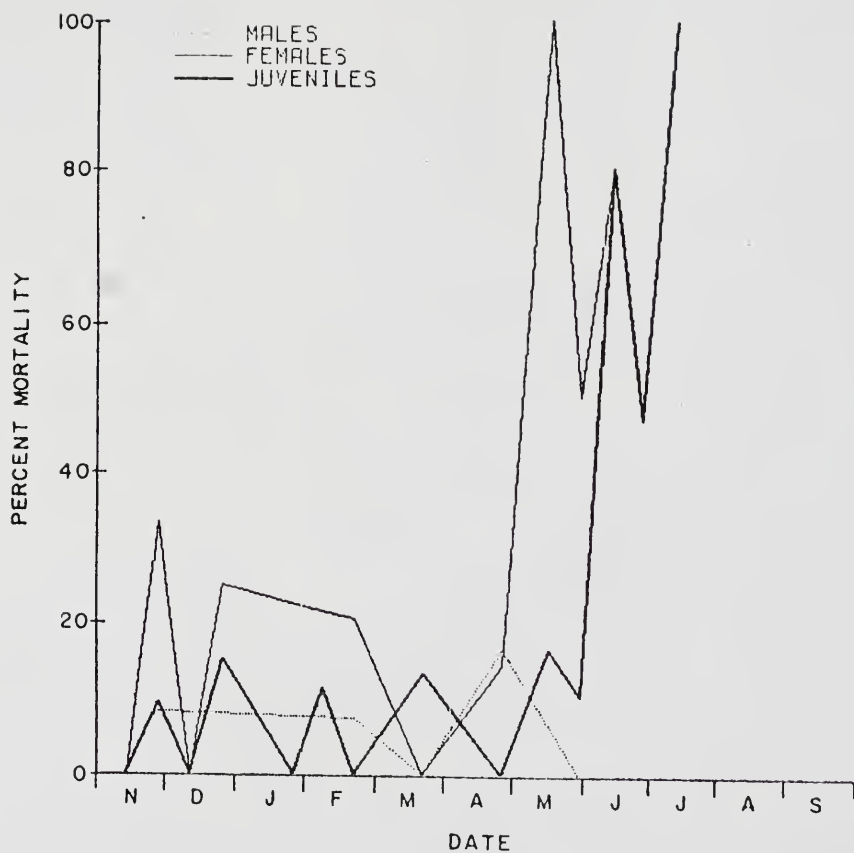


Fig. 30. Seasonal variation in mortality of *Euterpina acutifrons* exposed to treatment 102.

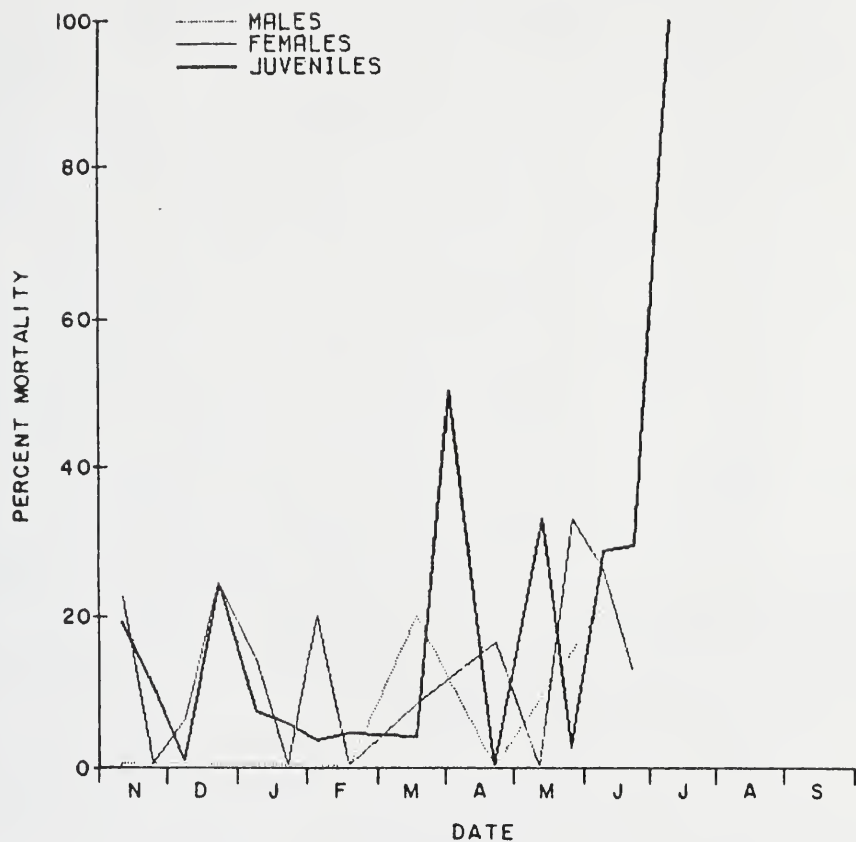


Fig. 31. Seasonal variation in mortality of *Euterpina acutifrons* exposed to treatment D50.

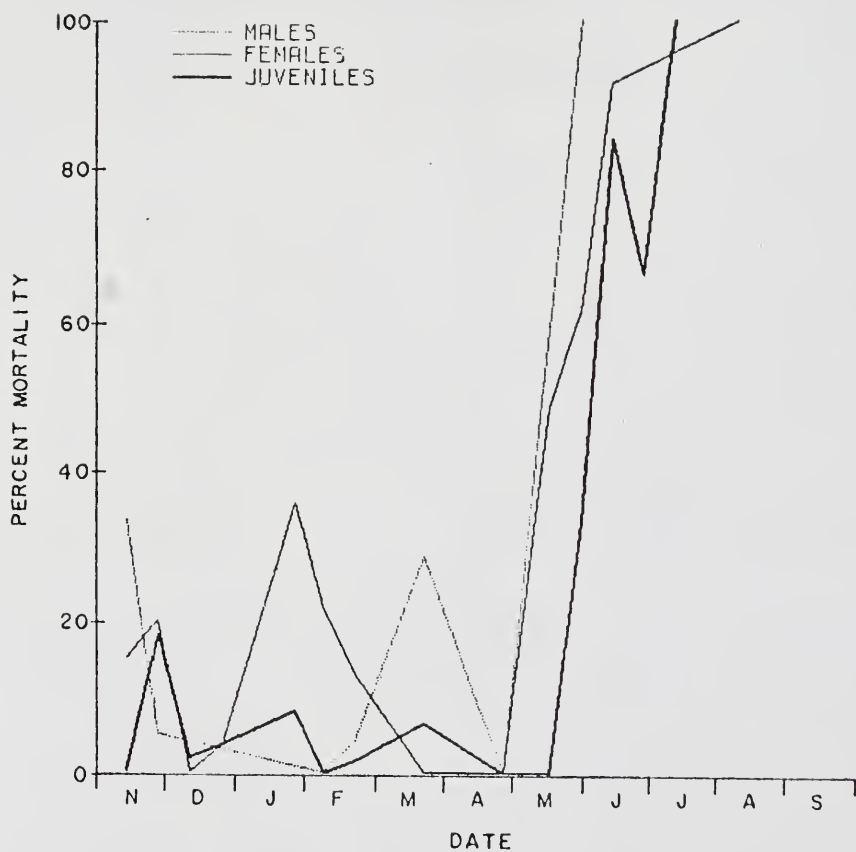
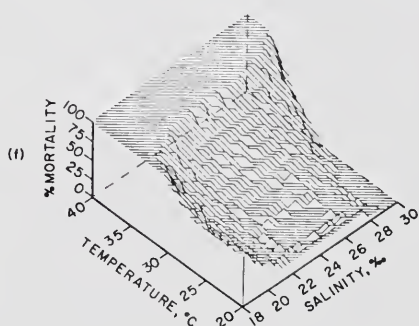
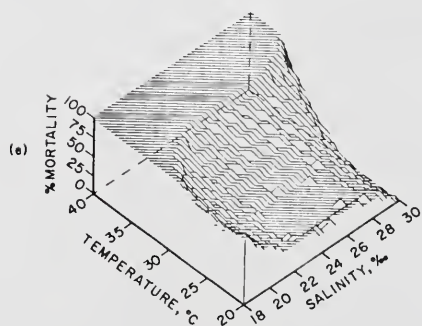
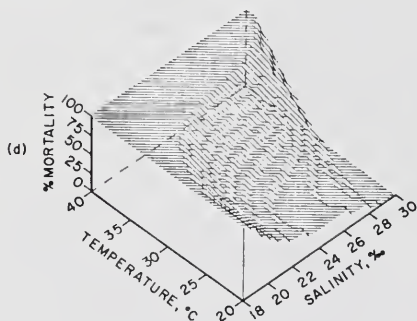
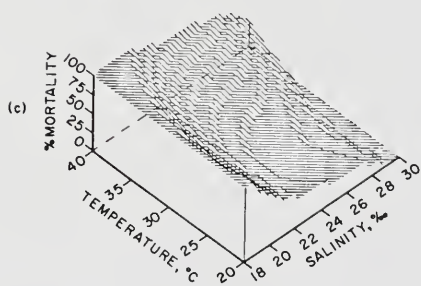
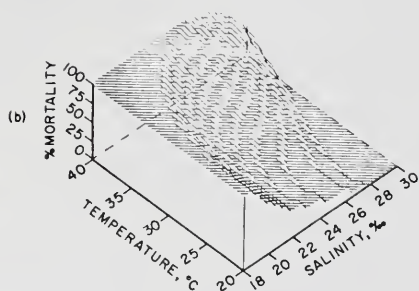
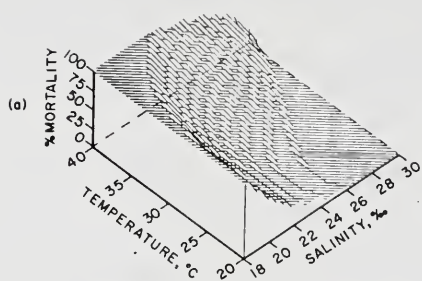


Fig. 32. Seasonal variation in mortality of Euterpina acutifrons exposed to treatment DS2.

Fig. 33. Response surface estimates of mortality as a function of discharge temperature and salinity for populations of Euterpina acutifrons exposed to various conditions: (a) male, (b) female and (c) juvenile immediate response to thermal shock; (d) female response to extended exposure to elevated temperatures; (e) female response to mechanical damage in addition to delayed thermal effects and (f) female response to same conditions in spring.



and only slightly increased with temperatures up to 30°C but then rose steeply to the 100% level by 35°C.

The predictive regression equation for Euterpina acutifrons was significant at the .0001 level ($F = 9.49$, 24df; $R^2 = .71$). The response surfaces that were plotted from the regression equation for various conditions are shown in Figure 33.

In response to short-term thermal stress, male Euterpina acutifrons (Figure 33a) are less sensitive to the various temperature and salinity combinations than are females (Figure 33b). Juveniles (Figure 33c) appear more resistant than adults, except for conditions of high salinity. The response surfaces for Euterpina acutifrons exhibit a stronger temperature-salinity interaction than do those of the other species. High salinities and low temperatures produce low mortalities, while low salinities and high temperatures produce high mortalities. The effects of this interaction on the immediate response to thermal shock appear more important than do either of the factors considered alone.

When discharge canal effects are considered (Figure 33d), mortalities rise, especially with high temperatures (above 35°C). Male Euterpina acutifrons have mortality levels 10 to 15% higher than the females, while juvenile mortalities are 10 to 15% lower. The temperature-salinity interaction remains evident but extended exposure to the heat makes the factor of temperature relatively more important in its upper range.

Mechanical effects cause mortality values in the response pattern for Euterpina acutifrons (Figure 33e) to rise slightly, the effect being especially evident in areas of low temperatures and high salinity

where mortalities were previously low. Most of the temperature-salinity interaction disappears and the temperature of the discharge canal becomes the dominant factor in entrainment mortality.

As with Paracalanus crassirostris, populations of Euterpina acutifrons entrained during the spring had significantly lower mortalities than would be expected (Figure 33f). Except at temperature above 35°C, mortalities are 10 to 20% lower throughout the range of conditions.

Euterpina acutifrons also showed response surface changes due to density factors. Conditions where this species had low numbers per cubic meter (Figure 26c) were related to higher mortalities, while those with high densities showed low mortalities (Figure 26d).

Pseudodiaptomus coronatus

Pseudodiaptomus coronatus is a fairly large calanoid copepod that is at times epibenthic. This species was the fifth most common copepod in the sampling area, but adults were quite rare. Male P. coronatus ranged in numbers from 0 to 41/m³ and had a mean value of 8/m³ (Figure 34). Females were not much more abundant, ranging in numbers from 0 to 47/m³ and having a mean value of 13/m³. Juveniles were at times quite common, however, ranging from 8/m³ to 934/m³ and having a mean value of 202/m³. Peak numbers occurred in December, March, June and September, though the winter peak was by far the greatest. The importance of this species, as indicated by the percent of the total population that it represented, fell from a high of 4% in December to a fraction of a percent by mid-spring (Figure 35).

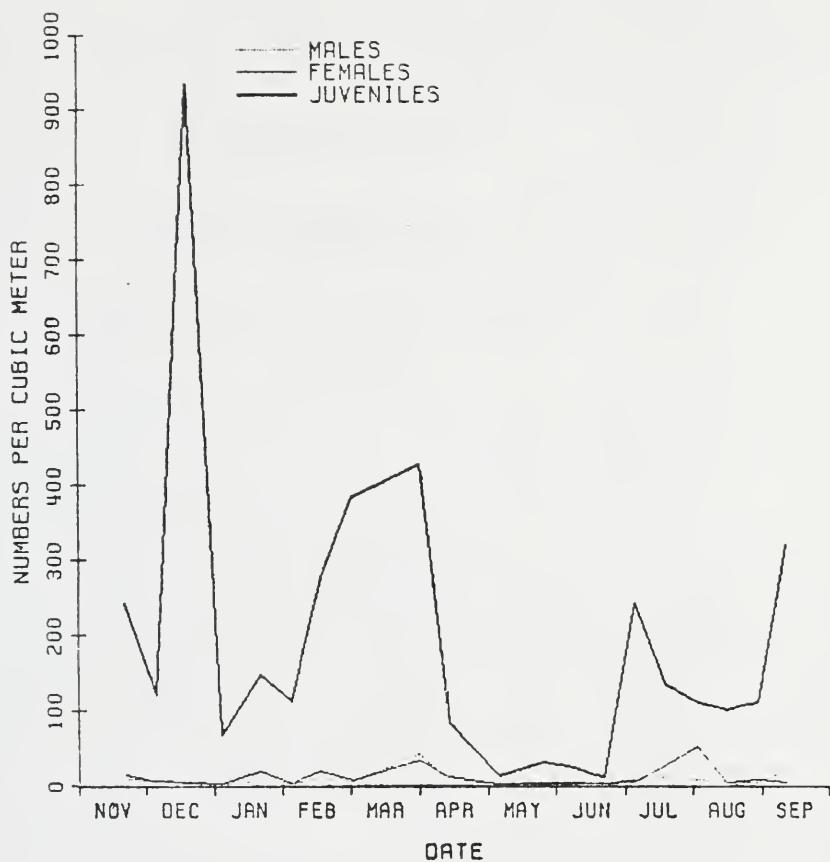


Fig. 34. Seasonal variation in numbers of Pseudodiaptomus coronatus.

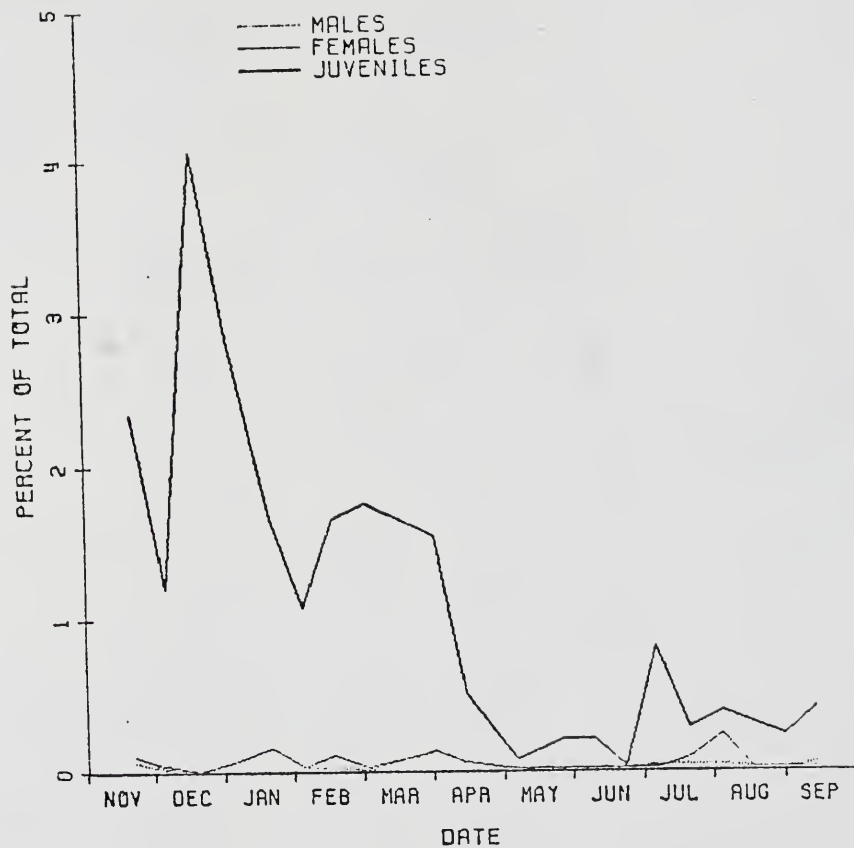


Fig. 35. Seasonal variation in percent of total copepod population represented by Pseudodiaptomus coronatus.

Pseudodiaptomus coronatus populations exposed to the thermal shock of discharge temperatures (treatment 1D0) exhibited a range of mortalities from 0 to 15% and had a mean value of 4%. The low numbers of this species in the control and experimental samples prevented mortality calculations for the adults subjected to this treatment. Juveniles exhibited low mortalities until mid August when this category also became too rare in the samples to permit further calculations (Figure 36).

Pseudodiaptomus coronatus subjected to treatment 1D2 exhibited mortalities from 0 to 100% with a mean value of 11%. Juvenile mortalities were the only values calculated for this treatment due to the scarcity of adults. Mortalities for the juveniles P. coronatus were low through the year until they suddenly rose to the 100% level in August (Figure 37).

Populations of Pseudodiaptomus coronatus sampled immediately following entrainment (treatment D50) had mortality values that ranged from 0 to 100% with a mean of 18%. Mortalities of juvenile P. coronatus fluctuated at moderately low levels through the year until August when values jumped to 100% (Figure 38). There were too few data points, for adult P. coronatus subjected to this treatment, to discern any trends.

The discharge population of Pseudodiaptomus coronatus that was left in the heated waters for two hours prior to sampling showed mortalities from 0 to 100% and a mean value of 23%. Juvenile P. coronatus had moderately low mortalities until the beginning of summer when values rose to higher levels (Figure 39). By September, mortalities for the juveniles jumped to the 100% level. Although there are only a few data points available for observation, female P. coronatus seem

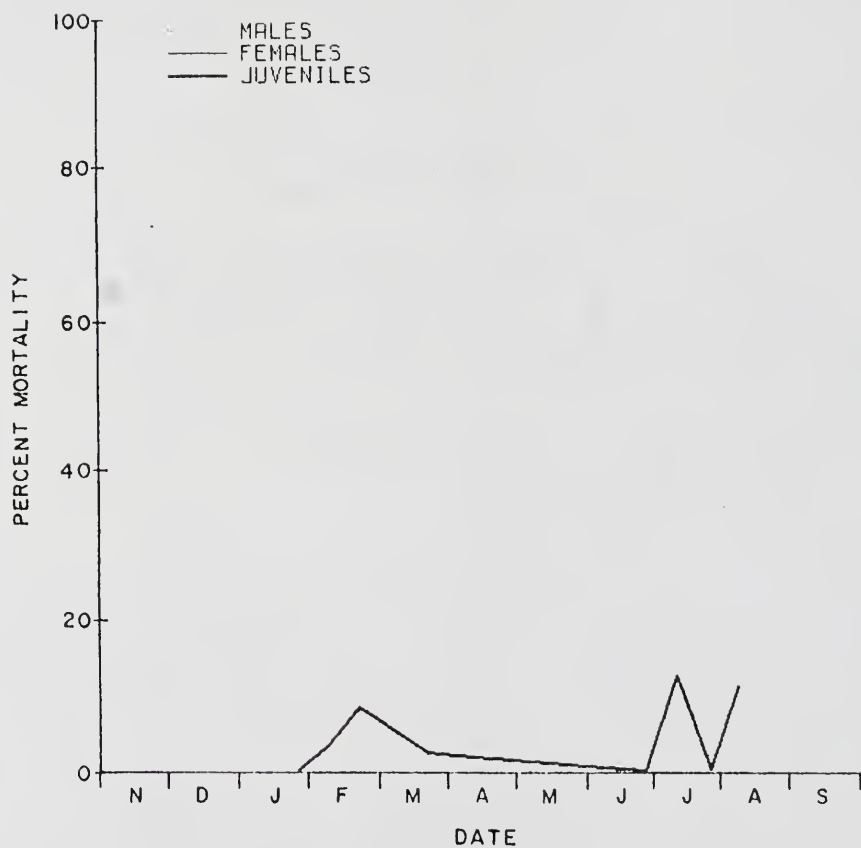


Fig. 36. Seasonal variation in mortality of *Pseudodiaptomus coronatus* exposed to treatment ID0.

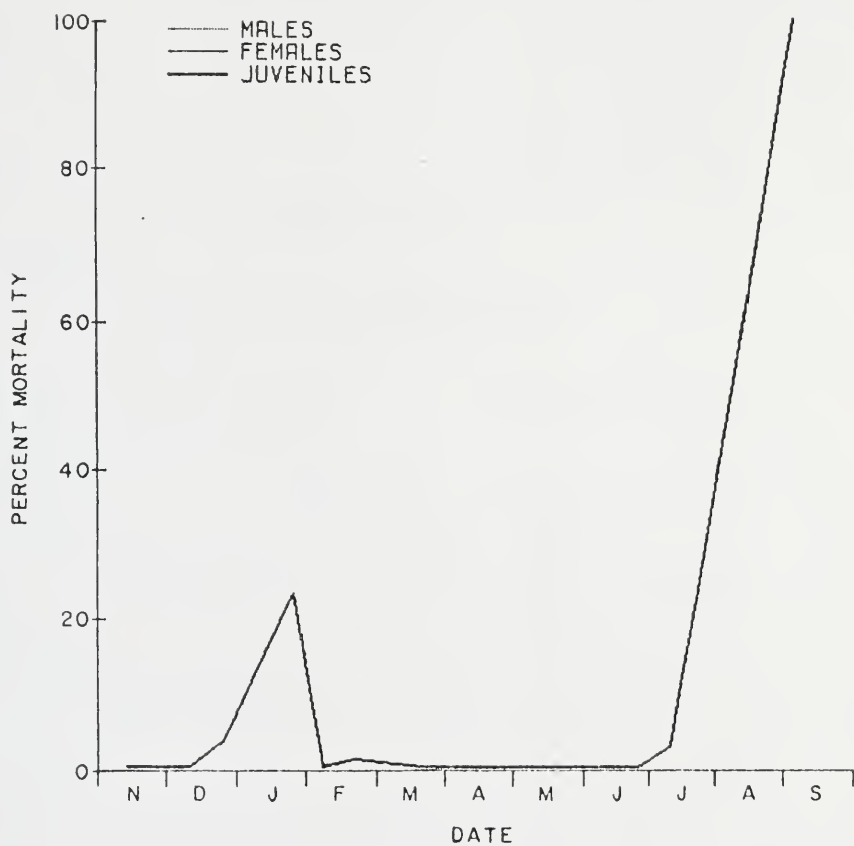


Fig. 37. Seasonal variation in mortality of Pseudodiaptomus coronatus exposed to treatment ID2.

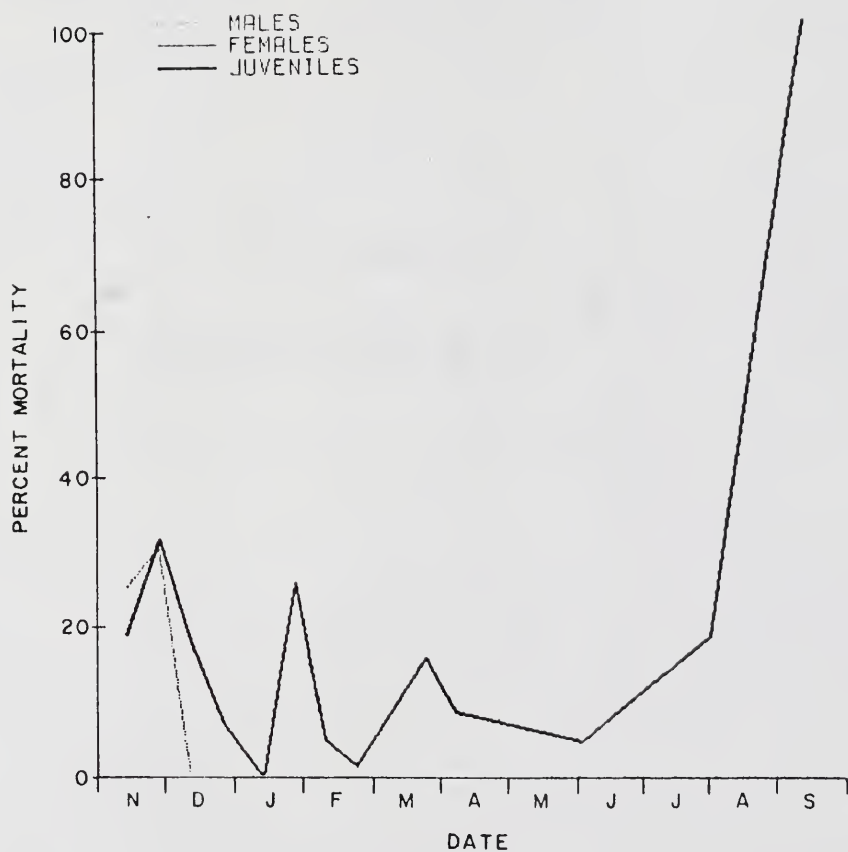


Fig. 38. Seasonal variation in mortality of Pseudodiaptomus coronatus exposed to treatment DS0.

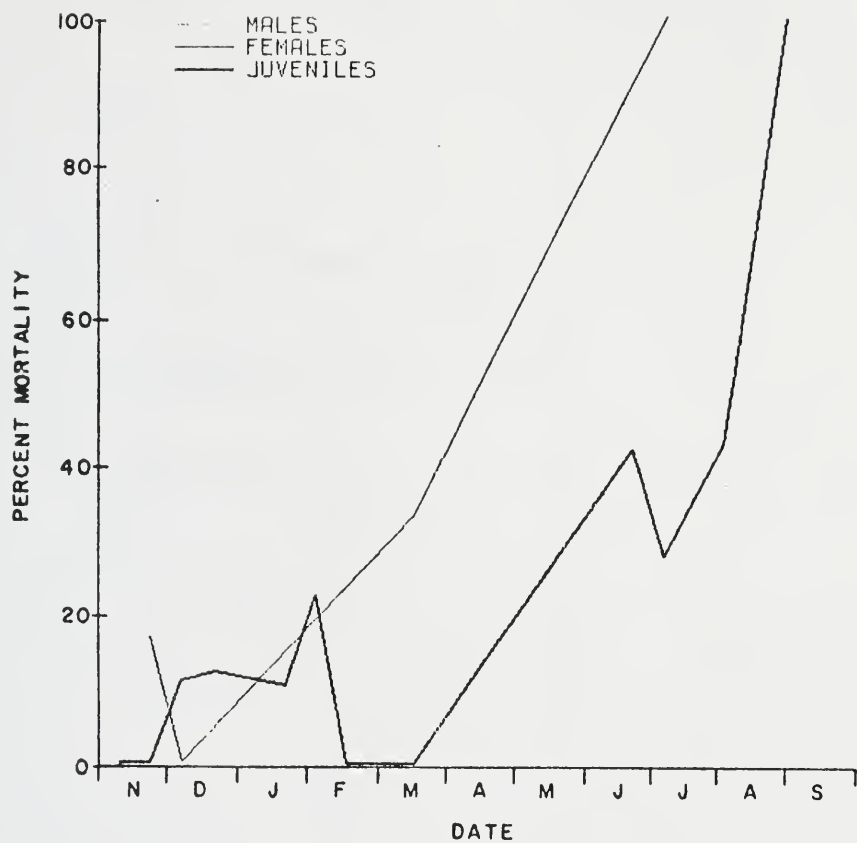


Fig. 39. Seasonal variation in mortality of *Pseudodiaptomus coronatus* exposed to treatment DS2.

to generally follow the same trend, though the 100% mortality level is reached in July.

Figure 10f shows the relationship between entrainment mortality of Pseudodiaptomus coronatus and the temperature of the discharge waters. Mortalities remained below the 20% level for observations below 35°C. Observations for temperatures around 35°C showed that mortalities had risen to the 40% level. By the time the temperature reached 37°C, mortalities went to 100%.

Multiple regression analysis and the creation of response surfaces were not attempted for Pseudodiaptomus coronatus due to the relatively low numbers of observations for each treatment. Such statistical treatment requires many observations for each of the various conditions in order to be considered valid.

Labidocera spp.

Labidocera spp. is a category made up of two species of the genus Labidocera: L. aestiva and L. scotti. These two species were counted together for two reasons. First of all, the numbers of either species alone were quite low, so that in order to obtain enough observations to analyze trends in mortality, the counts were combined. Secondly, although the species identification of these large calanoids was relatively easy as adults, the identification of juveniles to species was not feasible under the magnification used for counts. It was assumed that the effects of entrainment on the Labidocera species were more similar within the category than they would be to species of other genera.

Numbers of Labidocera spp. females ranged from 0 to $60/\text{m}^3$ with a mean of $8/\text{m}^3$ (Figure 40). Males of these species were rarer, ranging in numbers from 0 to $23/\text{m}^3$ and having a mean value of $3/\text{m}^3$. Juveniles were by far the most abundant, ranging from 0 to $567/\text{m}^3$ and averaging $25/\text{m}^3$. Peak numbers occurred in January, June and September.

Labidocera spp. represented between 0 and 2.7% of the copepod population of the sampling area, generally accounting for only about 0.2% of the total (Figure 41).

Populations of Labidocera spp. subjected to treatment ID0 exhibited mortality values that ranged from 0 to 40%, with a mean value of 13.2%. As can be seen in Figure 42, low numbers in experimental and control samples for this treatment allowed only a few data points to be plotted when the species are broken down by sex and age class. About the only trend that can be seen is that mortality values were fairly low for this treatment.

Labidocera spp. populations that were left in the heated effluent following thermal shock (treatment ID2) showed mortalities of 0 to 100% with a mean value of 49%. Mortalities were generally low until May (Figure 43). The coming of the warmer months brought consistently high mortalities except for a single dip in values at the end of June.

Discharge populations of Labidocera spp. sampled immediately following entrainment showed rather high mortality values ranging from 20 to 100% and having a mean value of 68%. Mortality values for adults were higher during the warmer months, remaining at the 100% level (Figure 44). Juvenile mortality fluctuated through the year, remaining at the 100% level only after mid-summer.

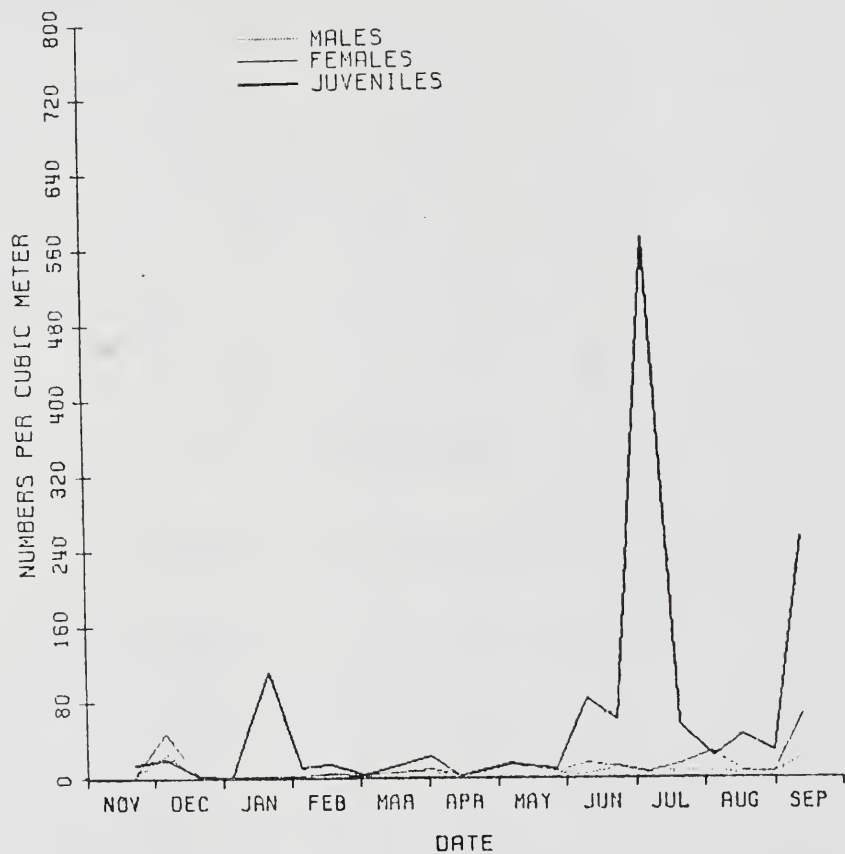


Fig. 40. Seasonal variation in numbers of Labidocera spp.

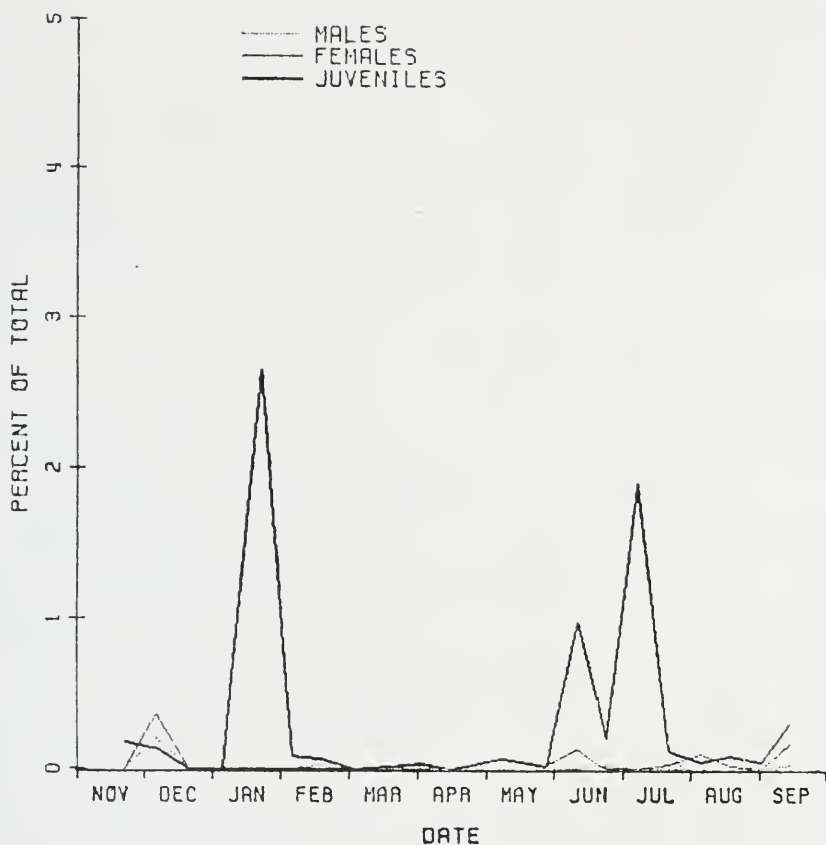


Fig. 41. Seasonal variation in percent of total copepod population represented by Labidocera spp.

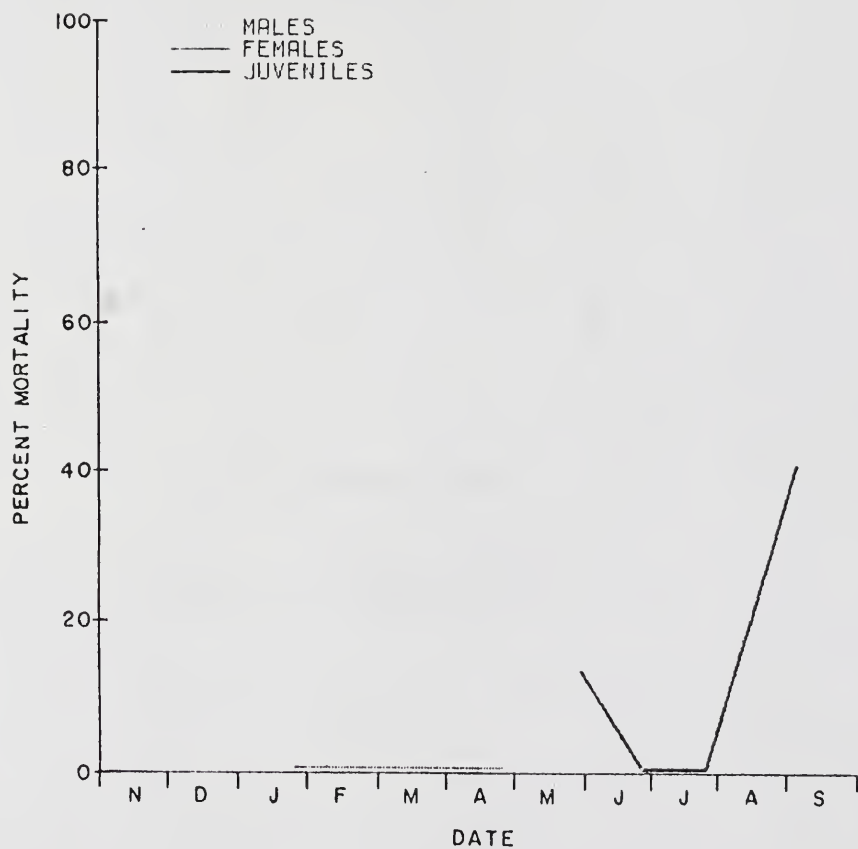


Fig. 42. Seasonal variation in mortality of *Labidocera* spp. exposed to treatment ID0.

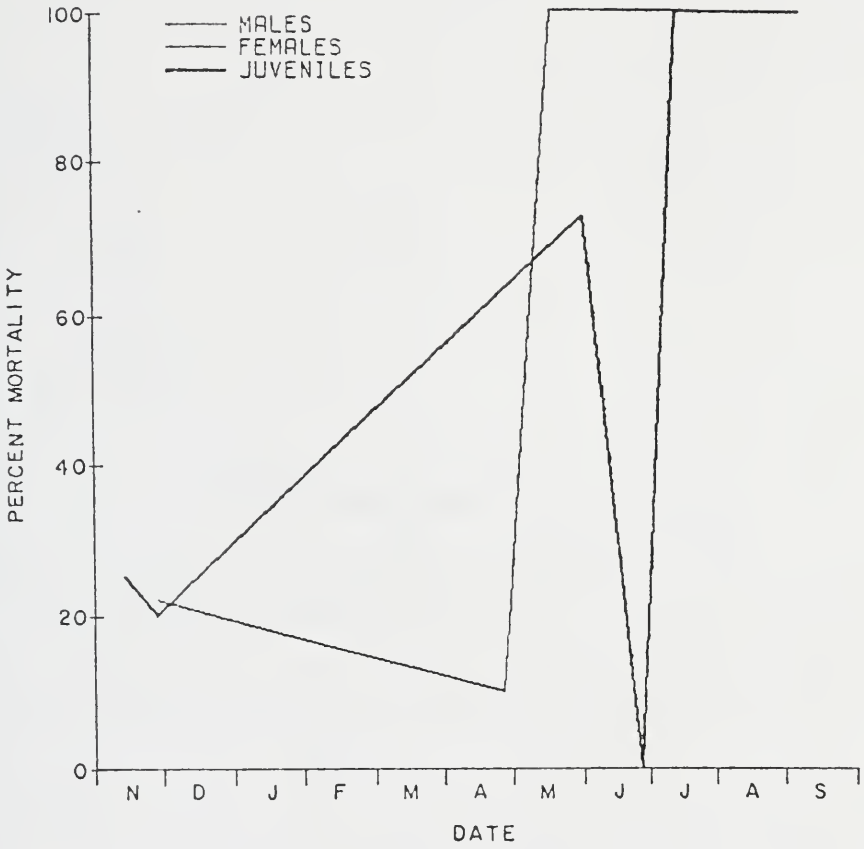


Fig. 43. Seasonal variation in mortality of *Labidocera* spp. exposed to treatment ID2.

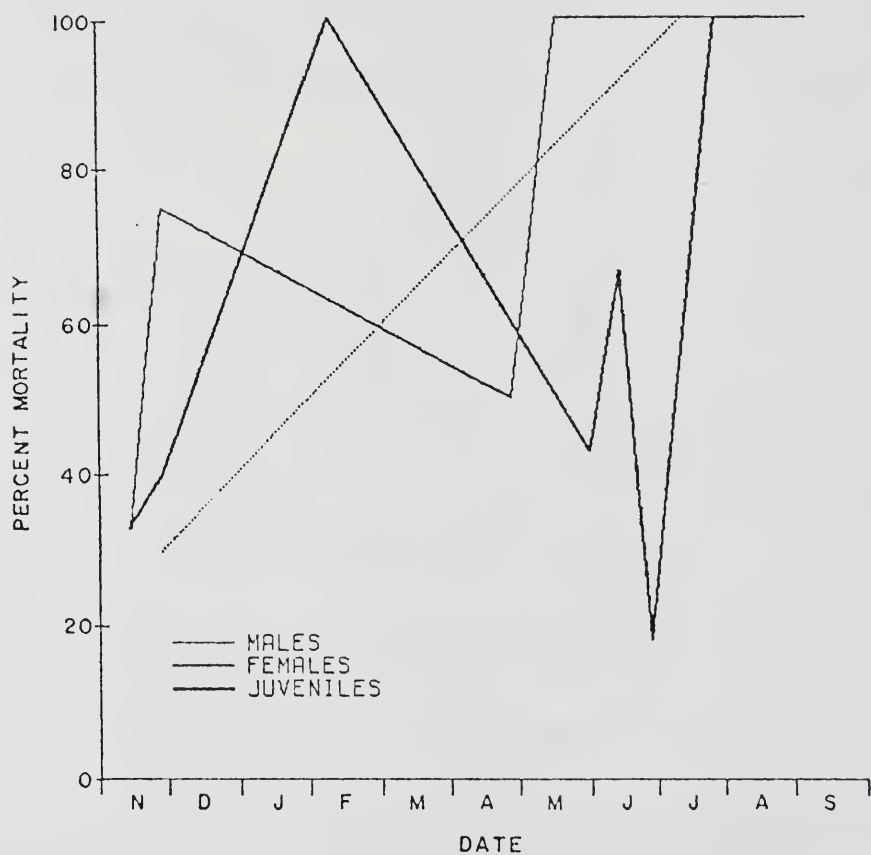


Fig. 44. Seasonal variation in mortality of *Labidocera* spp. exposed to treatment DS0.

Mortalities for Labidocera spp. subjected to treatment DS2 were high throughout the year, with a range of 75 to 100% and a mean value of 88%. Adult values fluctuated at high levels until May, then remained at the 100% level for the rest of the summer months (Figure 45). Juvenile mortalities showed greater fluctuations, but rose to 100% by mid-summer.

The relationship between the entrainment mortality of Labidocera spp. and discharge canal temperatures is shown in Figure 10e. Mortalities were high throughout the range of temperature, without an apparent trend. The only possible exception was that mortalities seemed to rise to higher levels and attain the 100% level more consistently with temperatures above 35°C.

Multiple regression analysis and the creation of response surfaces were not attempted for Labidocera spp. due to the low number of observations that could be made for the various conditions.

Tortanus setacaudatus

Tortanus setacaudatus is a fairly large calanoid copepod that was moderately abundant from time to time in the Crystal River area. Male T. setacaudatus ranged in numbers from 0 to 612/m³ and averaged 49/m³ (Figure 46). Females of this species numbered from 0 to 380/m³, with a mean value of 43/m³. Juveniles ranged in numbers from 0 to 845/m³ and had a mean of 127/m³. The major portion of the rather high mean numeric values of this species can be accounted for by three peak periods: one in April, one in August, and the last in September. During the April peak, this species represented 9% of the copepods of

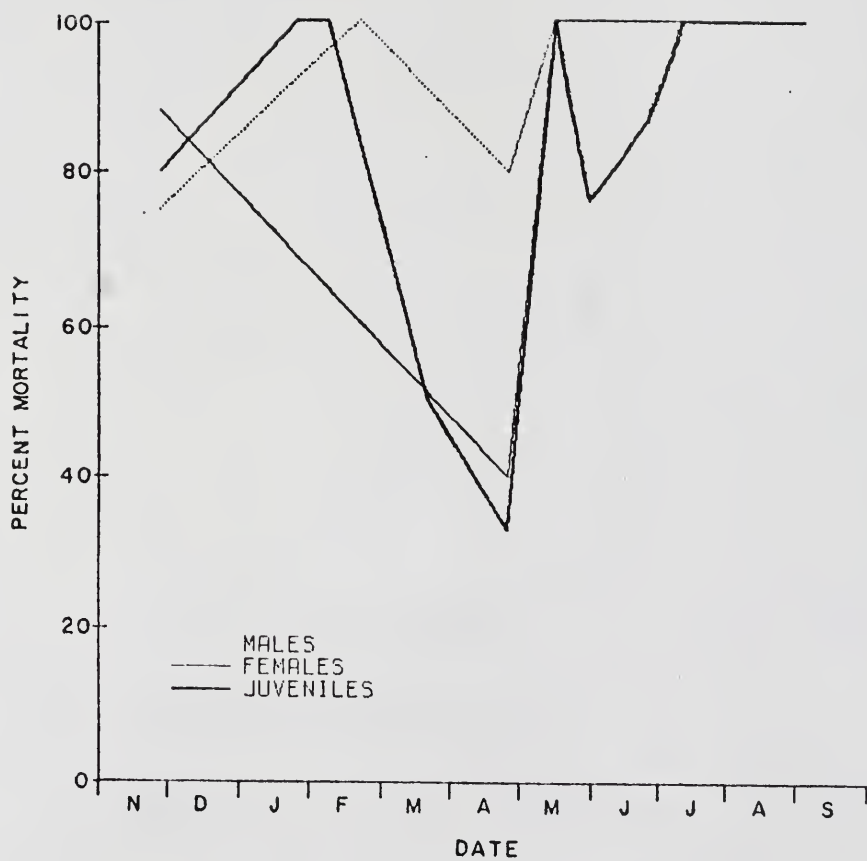


Fig. 45. Seasonal variation in mortality of *Labidocera* spp. exposed to treatment DS2.

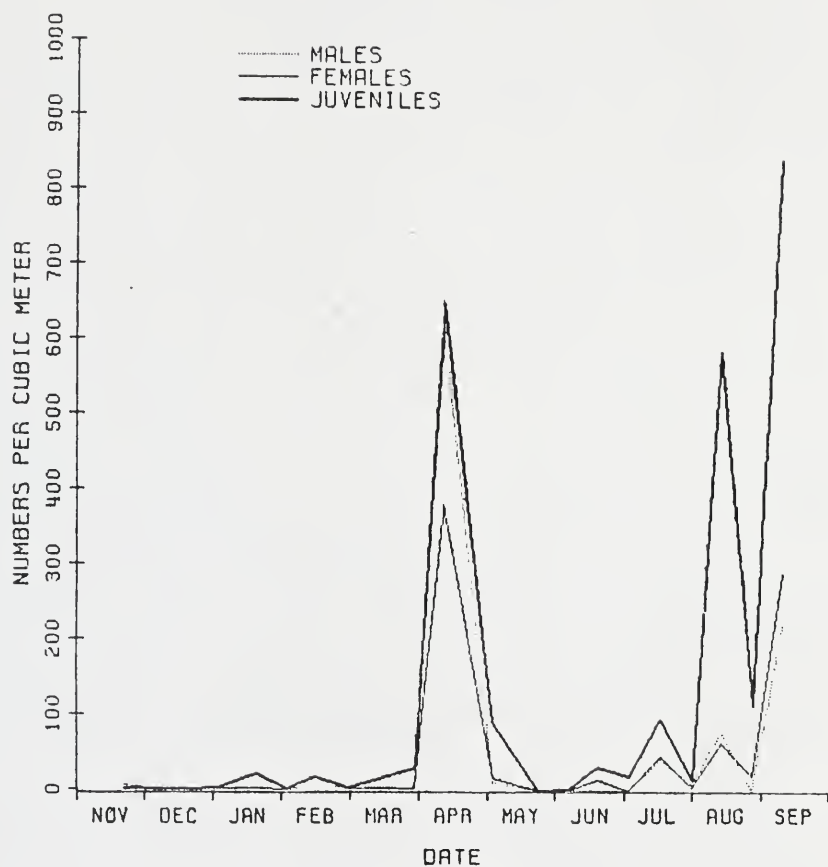


Fig. 46. Seasonal variation in numbers of *Tortanus setacaudatus*.

the area, while the August and September peaks accounted for approximately 1.8% of the total population (Figure 47). During the remainder of the year, however, T. setacaudatus comprised only a small fraction of a percent of the total copepod population.

Populations of Tortanus setacaudatus exposed to treatment ID0 had mortalities ranging in value from 0 to 100% with a mean of 19%. Male T. setacaudatus showed a single peak of 100% mortality in March and then dropped to very low levels for the remainder of the year (Figure 48): Females and juveniles exhibited low to moderate levels except for a peak at the 100% level at the end of August.

Tortanus setacaudatus populations that were allowed to remain in the effluent for several hours following thermal shock (treatment ID2) exhibited mortality values from 0 to 100%, with a mean of 31%. Mortalities were low through most of the year and rose to higher levels during the summer months (Figure 49). Juveniles tended to have the highest mortalities during this warmer period, while females had lowest mortalities.

Populations of Tortanus setacaudatus tested immediately following entrainment (treatment DS0) showed mortalities that ranged from 0 to 50% with a mean value of 8%. Mortalities for the species were generally low except for a peak of higher mortalities for the females in August (Figure 50).

The mortality values for the Tortanus setacaudatus population allowed to remain in the heated effluent following entrainment ranged from 0 to 100%, with a mean value of 27%. With the exception of a moderate peak in juvenile mortality in April, T. setacaudatus exhibited low mortalities until mid-summer when they rose sharply to the 100% level (Figure 51).

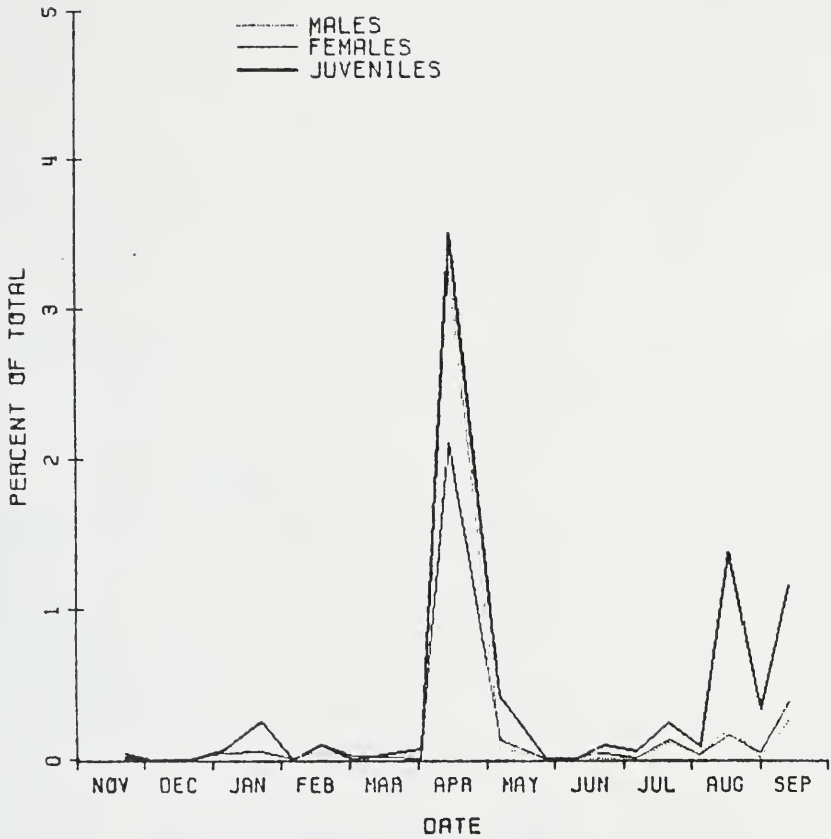


Fig. 47. Seasonal variation in percent of total copepod population represented by Tortanus setacaudatus.

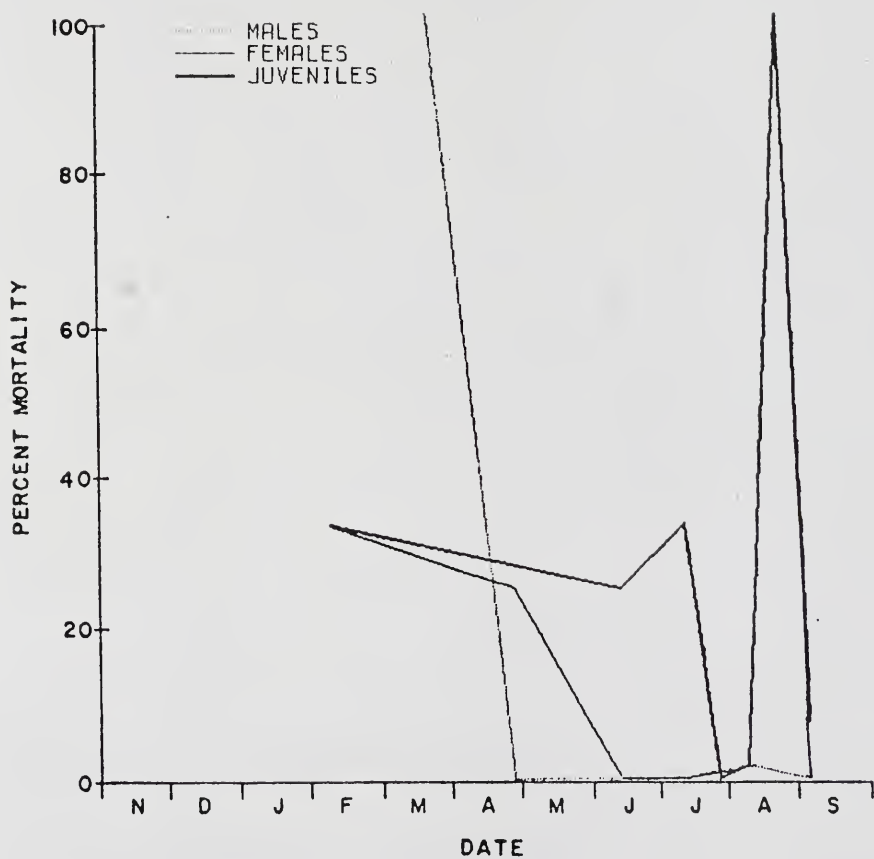


Fig. 48. Seasonal variation in mortality of *Tortanus setacaudatus* exposed to treatment ID0.

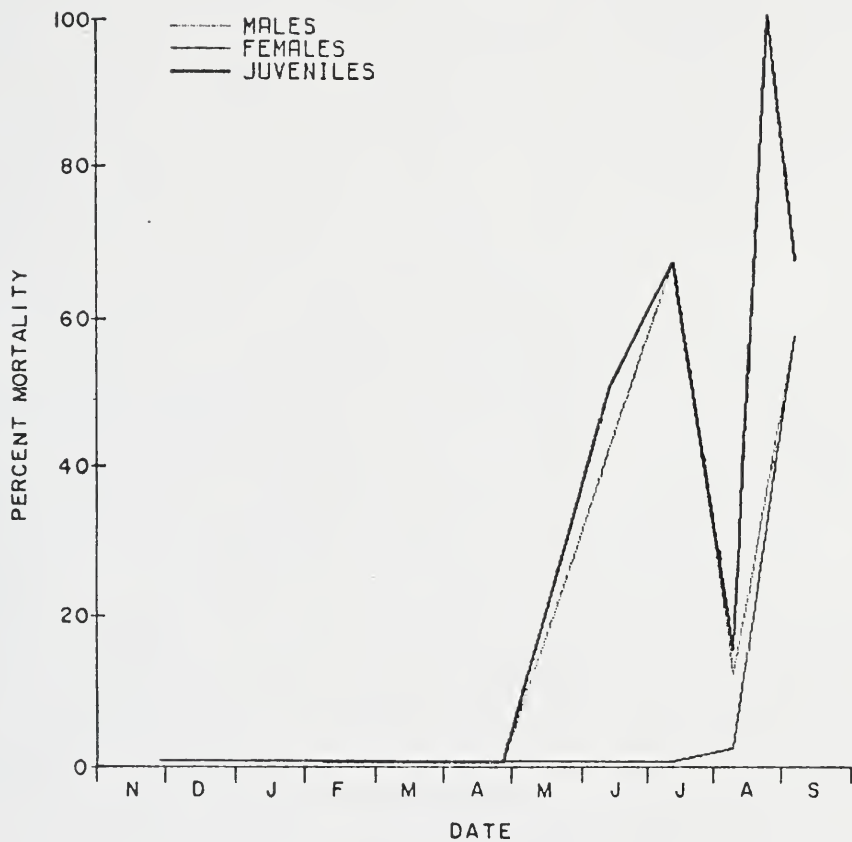


Fig. 49. Seasonal variation in mortality of *Tortanus setacaudatus* exposed to treatment ID2.

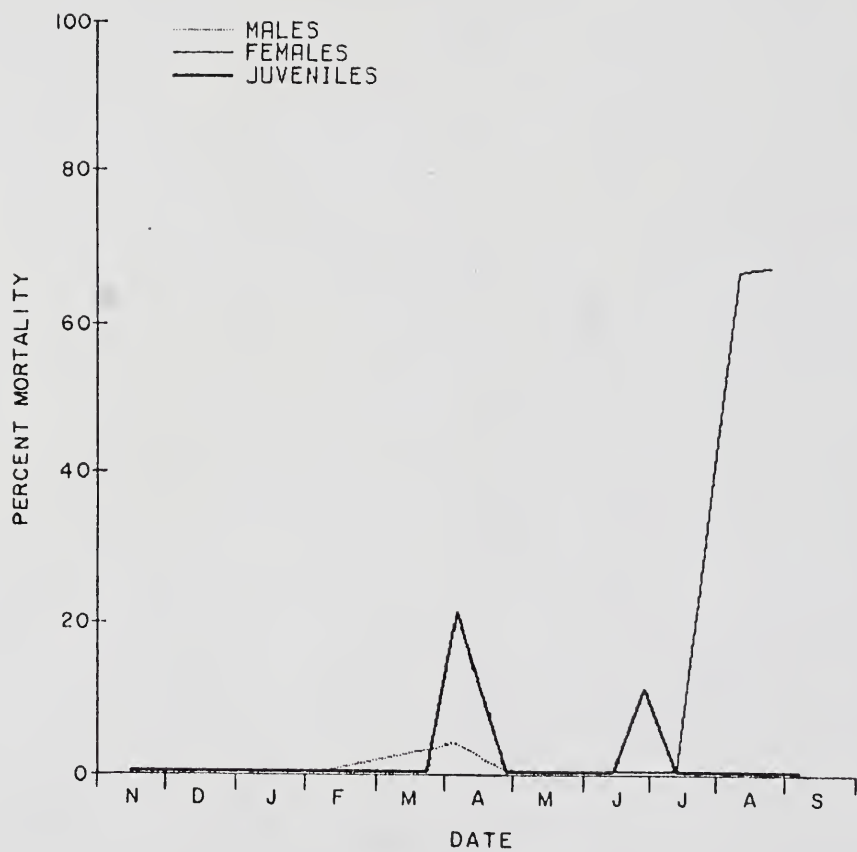


Fig. 50. Seasonal variation in mortality of *Tortanus setacaudatus* exposed to treatment DS0.

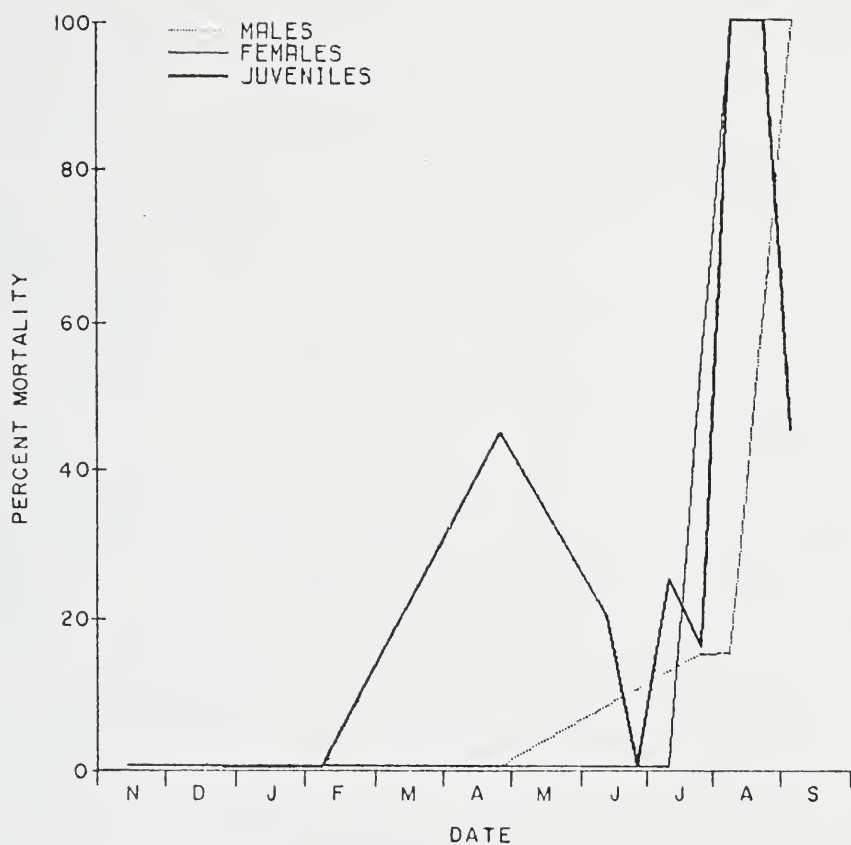


Fig. 51. Seasonal variation in mortality of *Tortanus setacaudatus* exposed to treatment DS2.

Figure 10g shows the relationship between the entrainment mortality of Tortanus setacaudatus and the temperature of the discharge waters. Mortalities were near 0% below 31°C, gradually increased to moderately low levels around 35°C and steeply rose as temperatures went above 36°C. The 100% mortality level was reached by this species only on the date that the discharge canal temperatures rose above 37°C.

Multiple regression analysis and creation of response surfaces were not attempted for Tortanus setacaudatus due to the low number of observations for the various conditions.

Total Copepod Population

Data from the copepod population as a whole basically reflect the trends of the dominant species, primarily Acartia tonsa and Oithona spp. An analysis of such data does, however, gives an overview of the general effects of entrainment on the copepods of the Crystal River area.

The number of female copepods sampled during the study ranged from 635/m³ to 22,791/m³, with a mean value of 7,233/m³ (Figure 52). Males were rarer, ranging from 106/m³ to 3,577/m³ and averaging 1,206/m³. Juvenile copepods were most abundant, ranging from 1,697/m³ to 44,452/m³ and having a mean value of 15,641/m³. Numbers generally rose from winter to summer, with peak numbers in December, March, July and September.

When the copepods were subjected to the thermal shock of treatment 1D0, mortalities ranged from 0 to 49% and averaged 8% over the course of the study. Mortality values were low through the winter and spring months, rose to slightly higher levels with the coming of summer, and

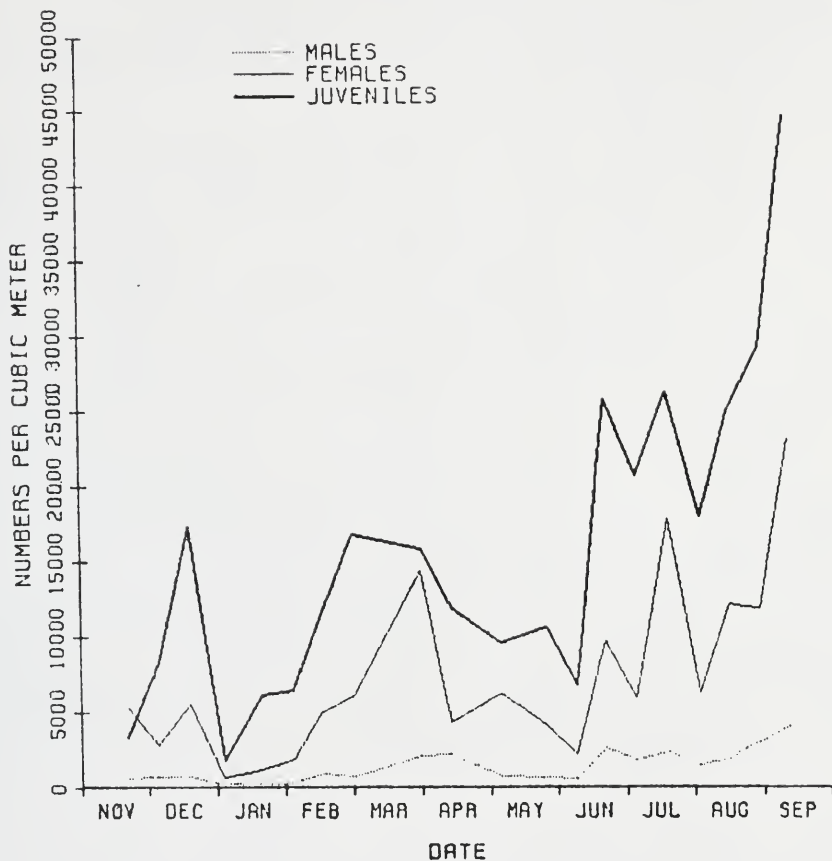


Fig. 52. Seasonal variation in numbers of the total copepod population.

peaked by the end of August (Figure 53). Males tended to have slightly higher mortalities during the warmer months.

Populations of copepods that were allowed to remain in the heated effluent following exposure to thermal shock (treatment 1D2) exhibited mortalities ranging from 0 to 73% and averaging 15%. Mortalities fluctuated at low levels until May, then gradually rose through July (Figure 54). By August, mortalities rose steeply to highest levels and remained there until the end of the study early in September. Males tended to have the highest mortalities during the critical summer months, while juveniles generally had the lowest values.

Copepods sampled for bioassay immediately following entrainment (treatment 1D0) showed mortalities from 0 to 46% with a mean value of 13%. With the exception of a single peak in April, copepod mortalities fluctuated at fairly low levels until mid-summer when they rose to higher levels (Figure 55). Male copepods appeared to be the most sensitive to this treatment throughout the year.

The DS2 treatment, which most closely simulates the conditions following entrainment, produced copepod mortality values that ranged from 1 to 77% with a year-round mean of 20%. Mortalities were moderately low through April, then rose throughout the remainder of the study (Figure 56). By the end of the summer, mortalities were at highest levels. Male copepods generally had higher mortality values than either females or juveniles.

The relationship between entrainment mortality of the copepods and discharge canal temperature is shown in Figure 10h. Mortalities were below 10% for temperatures less than 31°C. Above this temperature, mortalities rose at an increasingly rapid rate. The largest portion

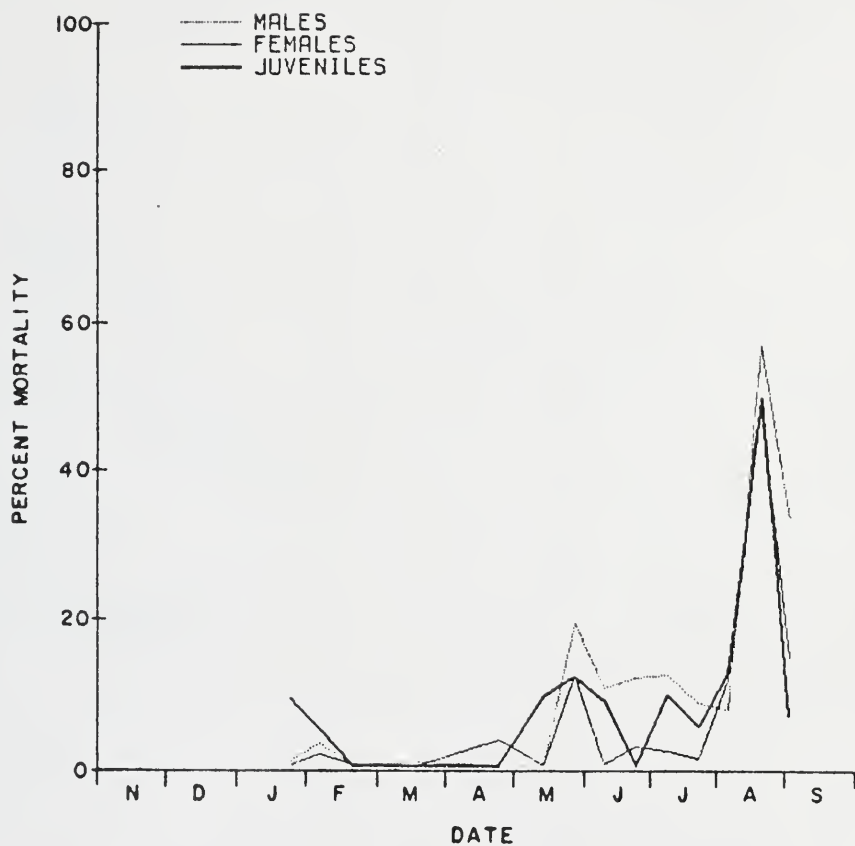


Fig. 53. Seasonal variation in mortality of the total copepod population exposed to treatment 1D0.

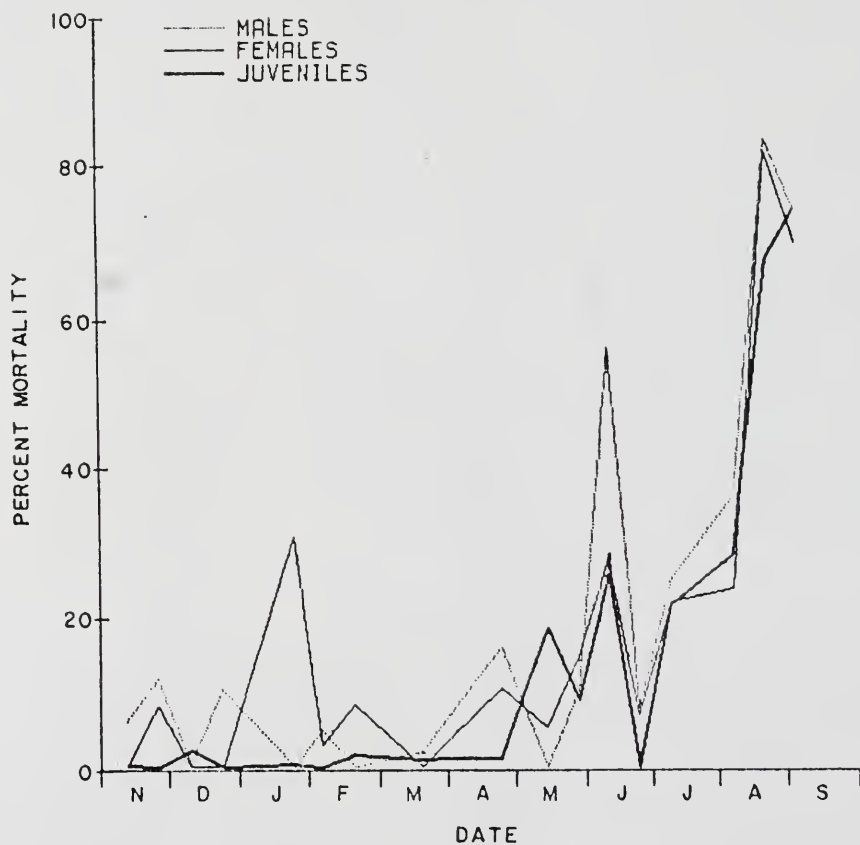


Fig. 54. Seasonal variation in mortality of the total copepod population exposed to treatment 102.

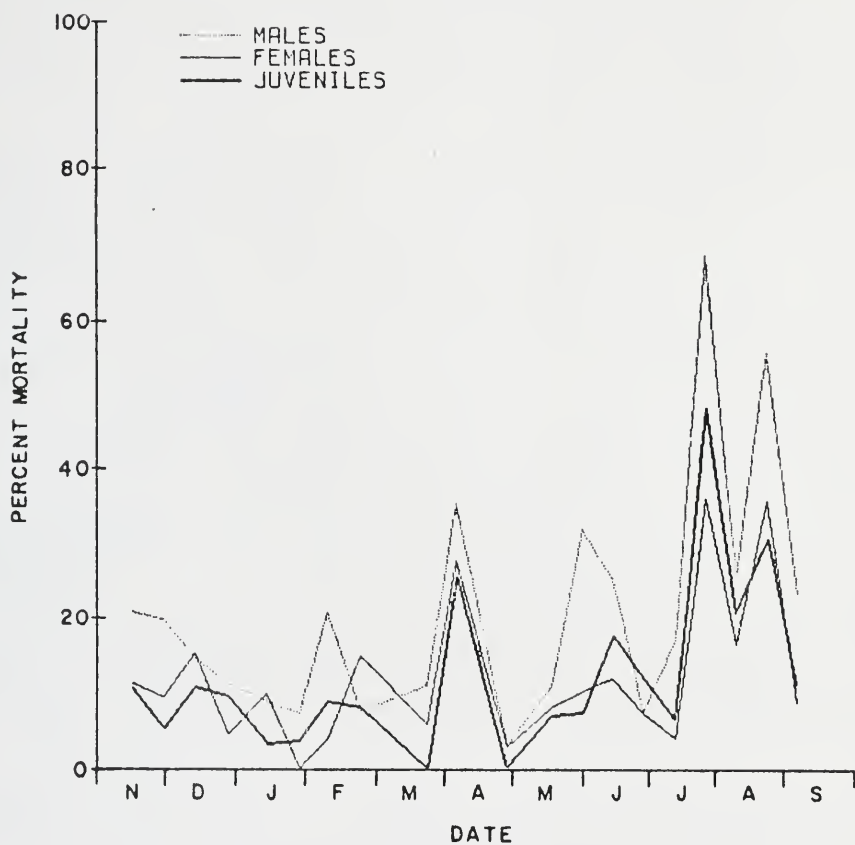


Fig. 55. Seasonal variation in mortality of the total copepod population exposed to treatment DS0.

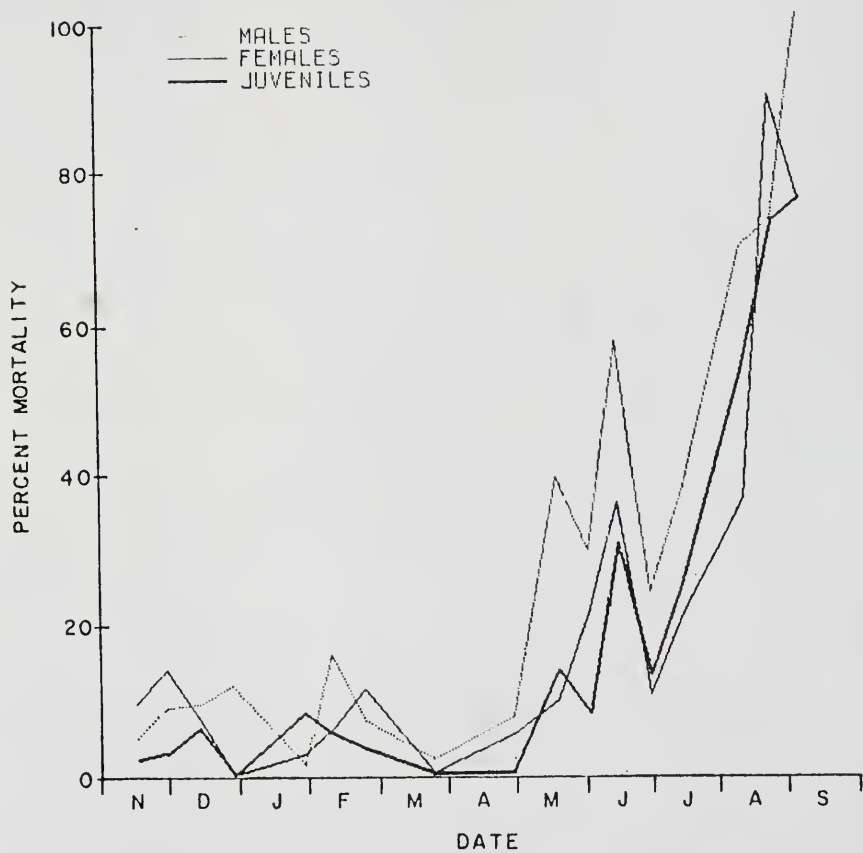


Fig. 56. Seasonal variation in mortality of the total copepod population exposed to treatment DS2.

of the increase in mortalities was seen between the temperatures of 35 and 37°C.

The predictive regression model for the total copepod population was significant at the .0001 level ($F = 61.2$, 16df; $R^2 = .84$). The response surfaces that were plotted from the regression equation for various conditions are shown in Figure 57.

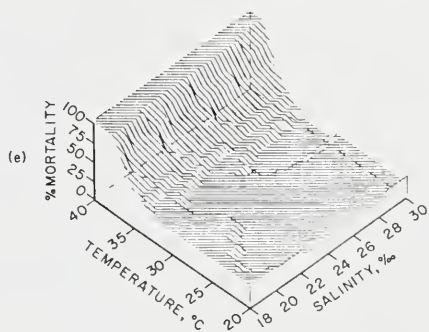
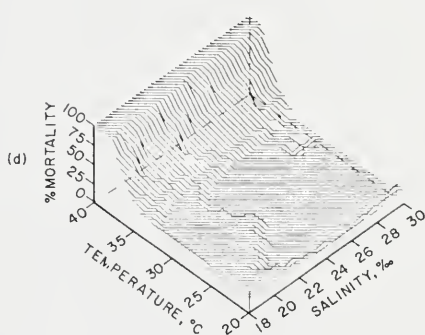
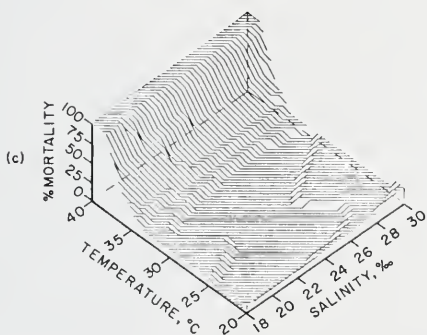
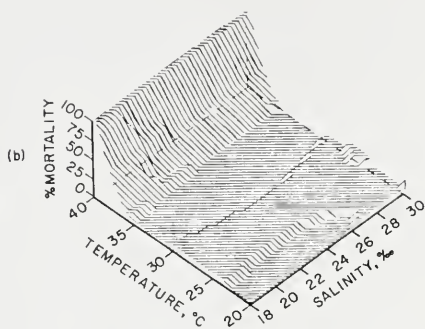
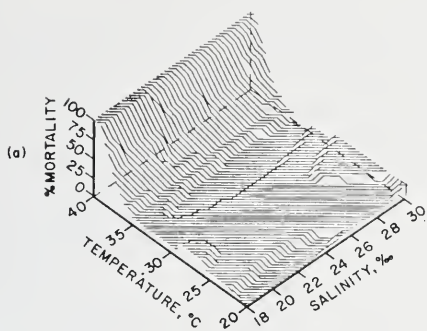
When subjected to thermal shock, male copepods (Figure 57a) exhibited slightly higher mortalities than females and juveniles (Figure 57b). Combinations of high salinity with moderately high temperatures appeared to produce slightly lower mortalities. Above 35°C, however, mortalities rose steeply across the entire range of salinity conditions.

Exposure to the heated waters of the discharge canal for several hours caused copepod mortalities to rise under most conditions (Figure 57c). Populations of copepods subjected to such stress under conditions of moderately high temperatures and high salinities or low temperatures and low salinities continued to exhibit lower mortalities. Copepods exposed to delayed thermal stress at temperatures above 35°C, however, appeared to be the most affected.

When mechanical effects are included in the entrainment conditions, copepod mortalities rose 10 to 20% (Figure 57d). Mortalities seen for copepods exposed to low salinity conditions were higher than those under higher salinity conditions.

The addition of seasonal effects associated with spring produced a response pattern for the copepods (Figure 57e) that looked identical to that for the other seasons (Figure 57d) except that all mortality values were approximately 6% higher.

Fig. 57. Response surface estimates of mortality as a function of discharge temperature and salinity for the total copepod population exposed to various conditions: (a) male, (b) female-juvenile immediate response to thermal shock; (c) female response to extended exposure to elevated temperatures; (d) female response to mechanical damage in addition to delayed thermal effects; and (e) female response to the same conditions in spring.



Laboratory Studies

Growth Studies of *Acartia tonsa*

Acartia tonsa juveniles grew well in culture and fairly large numbers of early stage nauplii could be collected from the sampling area throughout the year (Figure 58). The only exception was a period at the end of April and beginning of May when growth experiments had to be discontinued due to low numbers in the initial counts. The numbers of early stage A. tonsa nauplii basically followed the same trends as the numbers of adults and juveniles (Figure 4). The early instars (primarily naupliar stages 1 to 3) ranged in numbers from $312/\text{m}^3$ to $5,839/\text{m}^3$ with a mean of $1,701/\text{m}^3$. Peak numbers occurred in December, February, July and late August.

Growth curves created for each growth experiment are shown in Appendix A. Growth rates for intake and discharge populations on each sampling date were obtained by linear regression analysis of the growth curves and are shown in Figure 59. Pertinent information from the statistical comparison of the growth rates of control and entrained populations for each period is summarized in Table I. Growth rates for both populations of Acartia tonsa juveniles were similar in value until July (Figure 59). From July through the remainder of the summer the entrained populations exhibited growth rates that were consistently lower than those of control populations. The growth rates of A. tonsa populations from the intake area rose rapidly with the warmer months, while the rates for discharge populations during the summer tended to drop below previous levels. Differences between the growth rates of the populations during this period were statistically significant at

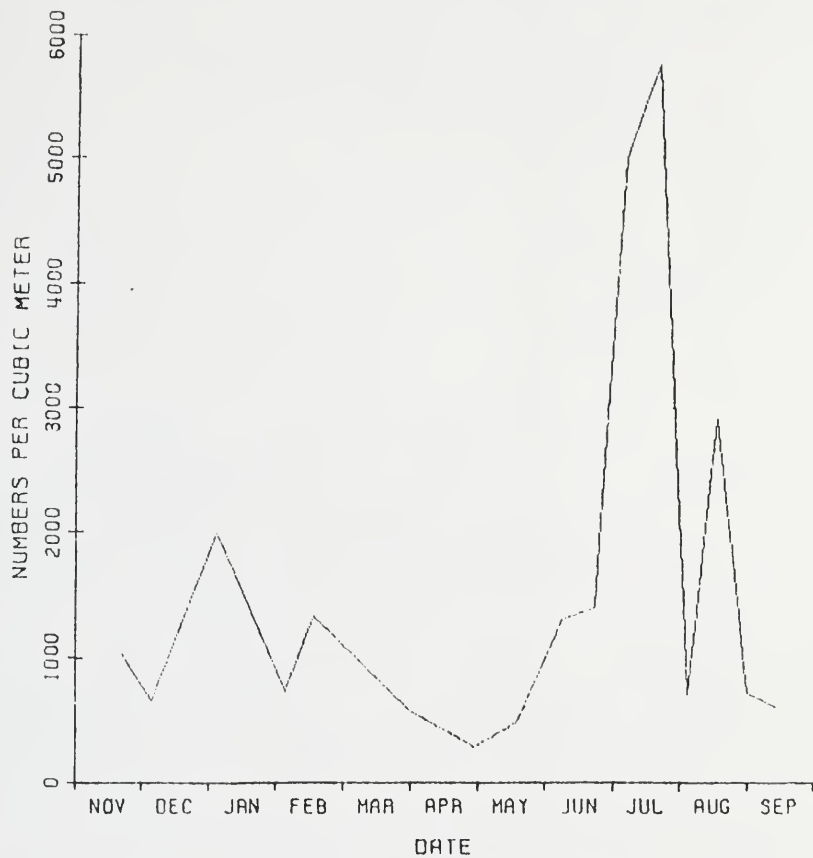


Fig. 58. Seasonal variation in numbers of early instars of *Acartia tonsa* found in samples at the beginning of growth experiments.

Fig. 59. Seasonal variation in growth rates of intake (closed circles) and discharge (open squares) populations of Acartia tonsa.

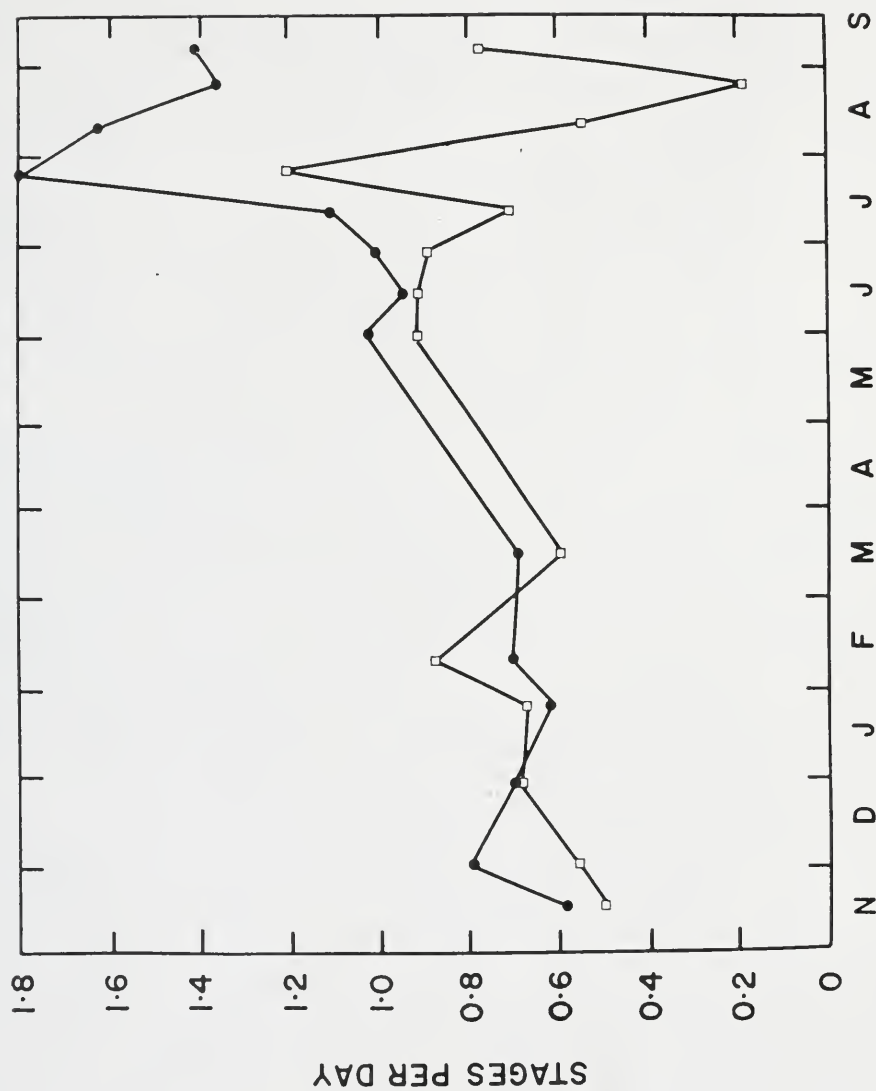


TABLE I
Comparison of the Growth Rates of Intake and Discharge *Acartia tonsa* Juveniles

DATE	TEMPERATURE (°C)		GROWTH RATES (stages/day)		.T-VALUE	LEVEL OF SIGNIFICANT DIFFERENCE
	Intake	Discharge	Intake	Discharge		
11-13-73	19.0	24.7	0.59	0.50	0.35, 6df	N.S.
11-27-73	23.8	29.5	0.79	0.54	1.27, 8df	N.S.
12-26-73	20.0*	21.5	0.69	0.68	0.02, 10df	N.S.
01-22-74	20.4	24.3	0.62	0.66	0.25, 12df	N.S.
02-05-74	21.0	25.8	0.69	0.88	0.77, 12df	N.S.
03-19-74	20.2	26.0	0.68	0.59	0.17, 7df	N.S.
05-28-74	27.5	34.5	1.19	0.91	0.58, 6df	N.S.
06-11-74	29.5	36.0	1.22	0.89	0.28, 3df	N.S.
06-25-74	27.2	34.2	1.03	0.87	0.55, 12df	N.S.
07-09-74	28.8	35.7	1.10	0.68	2.90, 11df	0.02
07-23-74	27.8	36.0	1.80	1.20	2.19, 12df	0.05
08-06-74	28.9	35.1	1.62	0.54	2.29, 10df	0.05
08-20-74	30.1	37.2	1.37	0.19	4.23, 10df	0.01
09-03-74	30.9	37.0	1.41	0.76	3.65, 10df	0.01

* Intake temperature of this date was 14°C but cultures were started at 20°C due to equipment failure.

the 0.05 level (Table 1). The average growth rate for control populations of A. tonsa over the period of significant differences was 1.46 stages/day, while that of entrained populations was 0.67 stages/day. The average difference of 0.79 stages/day between the growth rates of the two populations translates into an average depression in growth of 54% due to entrainment.

The relationship of temperature to entrainment effects on growth can be seen in Figure 60. The ratios of discharge to intake growth rates are plotted against discharge temperatures. Discharge growth rates were generally around 100% of control values for temperatures up to 30°C. For temperatures above this level, however, the ratio tended to decrease. As temperatures rose above 35°C, discharge growth rates were generally less than 60% of control values. Except for the sampling date in August when an unusually low discharge to intake growth rate ratio was shown for a temperature of 35.1°C, the values of the ratio tended to fall rapidly with increasing temperatures.

The general shape of the curve traced by the growth rate ratios appears to be a horizontal line for most of the temperature range, followed by an exponential decline with highest temperatures. This type of curve might be seen as the inverse of the curve that describes the mortalities observed for most species in Figure 10.

To better observe what is happening within the Acartia tonsa populations during the period of growth depression, age structure histograms were created for the two populations on each day that counts were made. The resulting series of histograms showed population structure changes throughout each experimental period. A representative series of the histograms from the period of significant growth depression is

Fig. 60. Growth rate ratio of Acartia tonsa as a function of discharge temperature. For temperatures with more than one observation, the vertical line represents one standard error above and below the mean value.

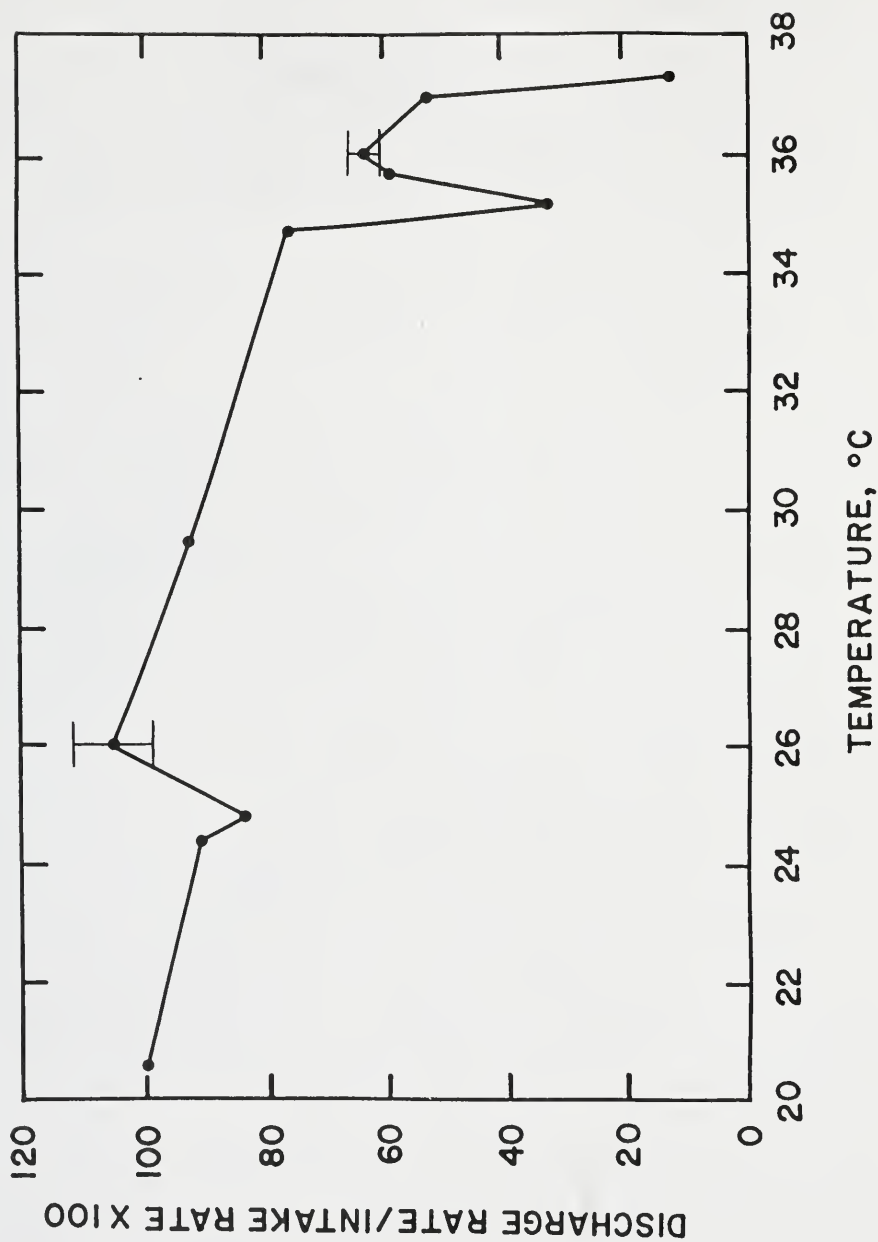
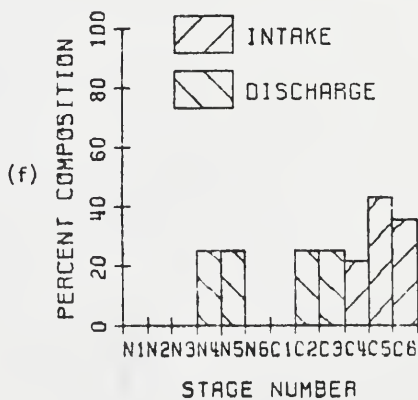
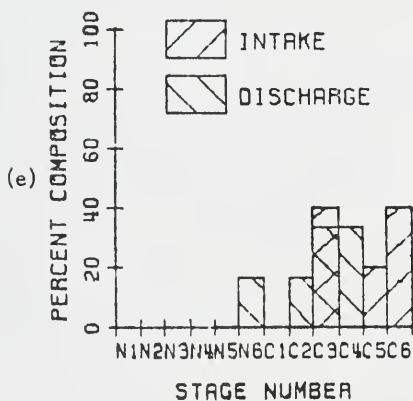
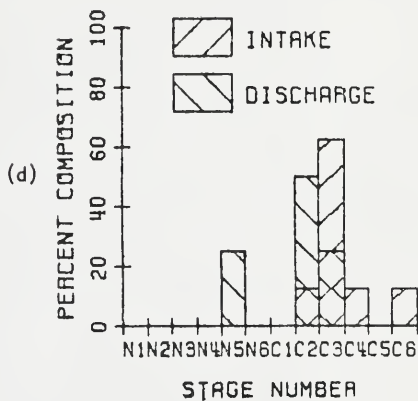
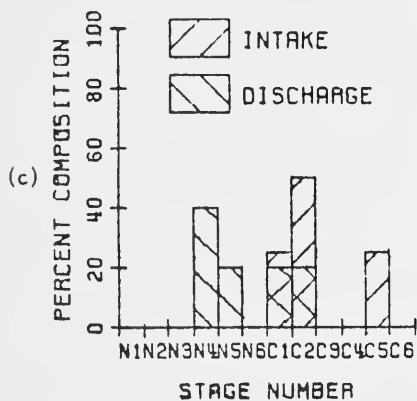
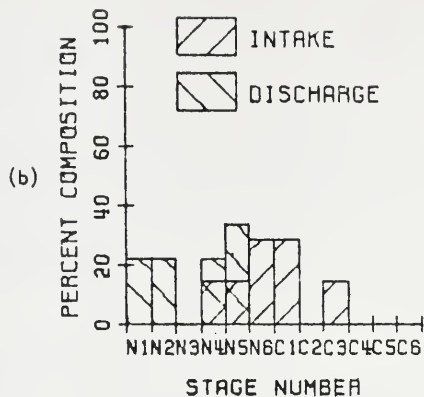
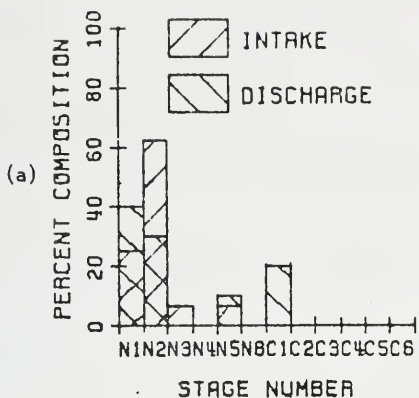


Fig. 61. Histogram series for Acartia tonsa in growth experiment started on September 3, 1974, showing changes in population structure with time: (a) Day 0.3. (b) Day 3, (c) Day 4, (d) Day 5, (e) Day 6, (f) Day 7. N1 to N6 are the six naupliar stages, while C1 to C5 are the five copepodite stages. C6 is the adult.



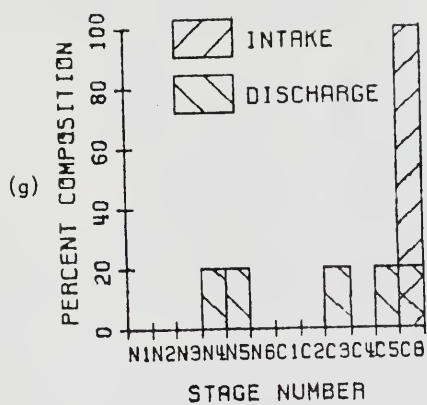


Fig. 61. Continued
(g) Day 8

shown in Figure 61, while histogram series for experiments throughout the remainder of the year are shown in Appendix B. As one follows the histograms in Figure 61 through the days of the growth experiment, the age structure of the intake population shifts consistently towards the older stages until, by day 8, all of the population has grown to adulthood (Stage C-6). The discharge population, on the other hand, has a portion of the population remaining as early instars even though the rest of the population grows to older stages. On day 8, 40% of the population were still in naupliar stages even though the rest of the population approached adulthood. This trend for some of the population to live without apparent growth was seen for all experimental periods where there was significant growth depression. In addition, the growing portion of affected populations generally appeared to lag behind the controls in growth.

Growth Studies of *Oithona brevicornis*

Oithona brevicornis juveniles also grew well in culture and early stages of this species were always a dominant portion of the microplankton of the sampling area (Figure 62). Numbers of the early instars ranged from $1,955/m^3$ to $53,906/m^3$ and had an average value of $14,792/m^3$. Peak numbers occurred at the end of May and the general trend showed density of juveniles to increase from winter to summer.

The growth curves produced by the growth experiments on *Oithona brevicornis* are shown in Appendix A. Figure 63 shows the relationship between growth rates calculated for entrained and control populations throughout the year. Growth rates of *O. brevicornis* juveniles were

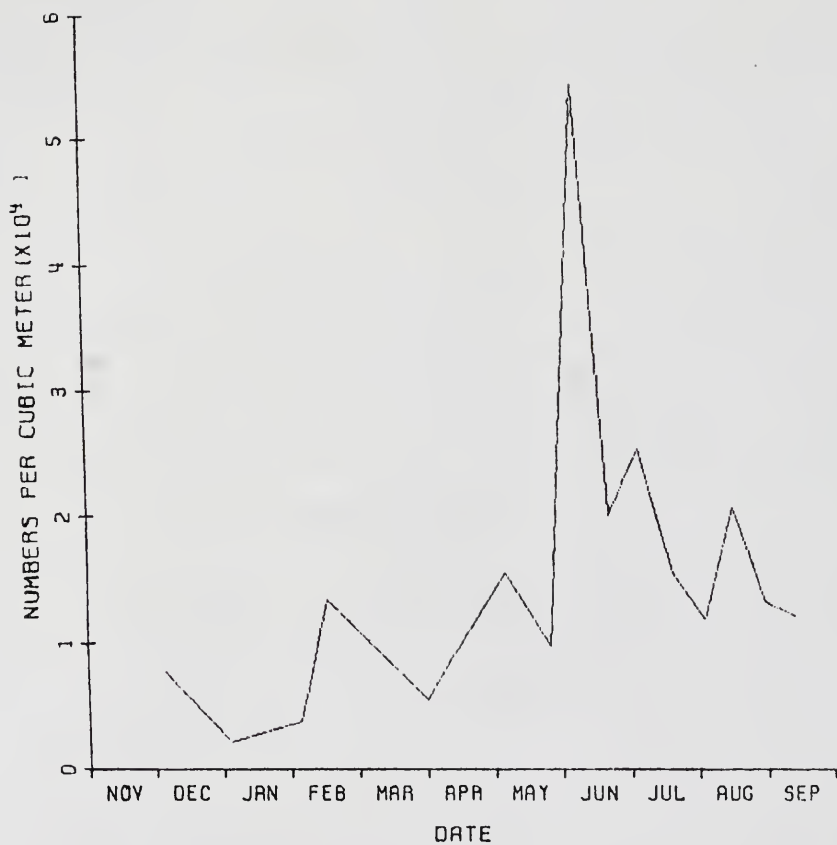
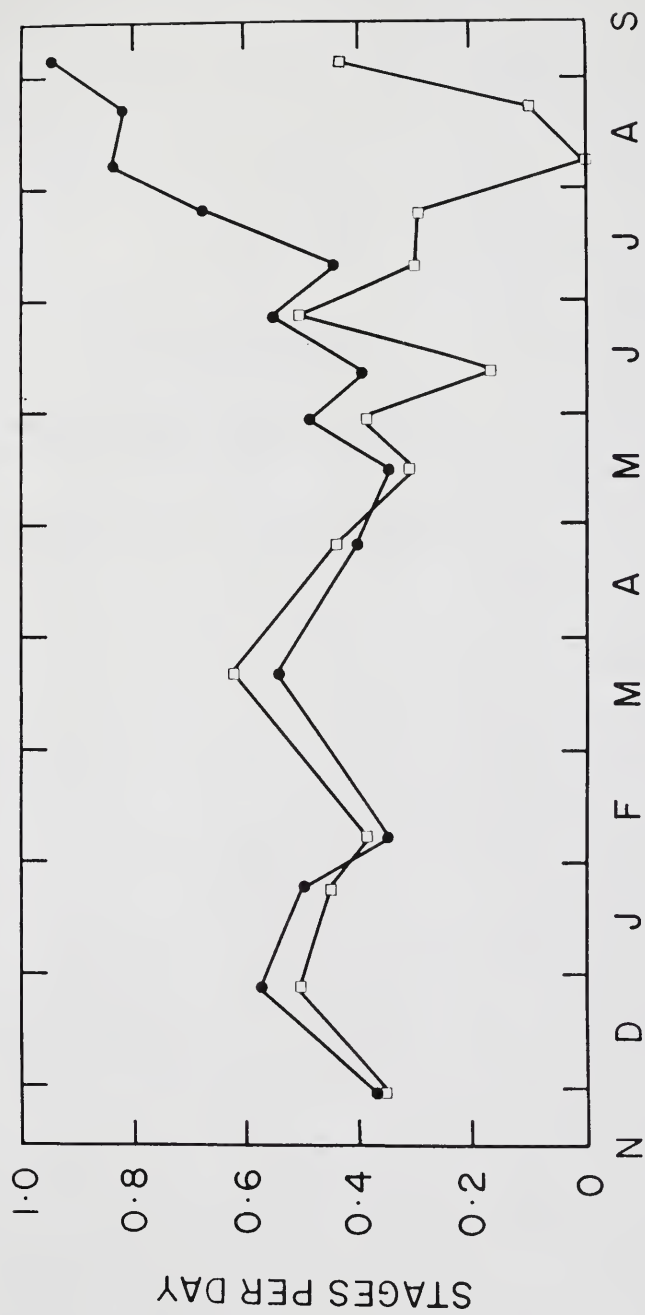


Fig. 62. Seasonal variation in numbers of early instars of *Oithona brevicornis* found in samples at the beginning of growth experiments.

Fig. 63. Seasonal variation in growth rates of intake (closed circles) and discharge (open squares) population of Oithona brevicornis.



much slower than those found for Acartia tonsa. For the major portion of the year, control populations grew at a little less than 0.5 stages/day. As temperatures rose in the summer, the growth rates for the control organisms increased rapidly to nearly 1 stage/day by the end of the summer. The entrained population exhibited growth rates similar to control values until mid-summer. From the middle of July to the end of the study in early September, however, discharge populations of O. brevicornis juveniles exhibited significant depression of growth rates (Table 2). The average growth rate for control populations during this critical period was 0.81 stages/day while that of entrained juveniles was only 0.20 stages/day. The 0.61 stages/day difference between the average intake and discharge growth rates represents an average depression of growth of just over 75%.

The relationship between temperature and entrainment effects on growth are seen in Figure 64. The growth rates of Oithona brevicornis juveniles from the discharge area were close to 100% of control values throughout most of the range of temperatures. Above 35°C, however, the ratio of discharge to intake growth rates drops to low levels. As was seen for Acartia tonsa, the growth rate ratio was extremely low for the August sampling period when temperatures registered only 35.1°C. The general trend seems to show little entrainment effect for most temperatures, but an exponential decline in growth rate ratio values as temperatures exceeded 35°C.

Series of age structure histograms were created for the growth experiments on Oithona brevicornis and are shown in Appendix B. A representative series of histograms for an experiment during the period of growth depression is shown in Figure 65. Intake populations

TABLE 2
Comparison of the Growth Rates of Intake and Discharge *Oithona brevicornis* Juveniles

DATE	TEMPERATURE		GROWTH RATES (stages/day)		T-VALUE	LEVEL OF SIGNIFICANT DIFFERENCE
	Intake	Discharge	Intake	Discharge		
11-27-73	23.8	29.5	0.36	0.34	0.11, 12df	N.S.
12-26-73	20.0*	21.5	0.57	0.51	0.17, 8df	N.S.
01-22-74	20.4	24.3	0.50	0.44	0.26, 12df	N.S.
02-05-74	21.0	25.8	0.34	0.37	0.06, 6df	N.S.
03-19-74	20.2	26.0	0.55	0.62	0.31, 12df	N.S.
04-24-74	23.9	30.6	0.39	0.42	0.18, 12df	N.S.
05-14-74	25.1	31.3	0.34	0.30	0.27, 10df	N.S.
05-28-74	27.5	34.8	0.48	0.38	0.83, 12df	N.S.
06-11-74	29.5	36.0	0.45	0.14	0.69, 5df	N.S.
06-25-74	27.2	34.4	0.55	0.49	0.29, 12df	N.S.
07-09-74	28.8	35.7	0.43	0.29	0.78, 12df	N.S.
07-23-74	27.8	36.0	0.66	0.28	2.21, 12df	0.05
08-06-74	28.9	35.1	0.82	0.00	4.00, 12df	0.01
08-20-74	30.1	37.2	0.81	0.08	4.29, 12df	0.01
09-03-74	30.9	37.0	0.93	0.43	3.22, 10df	0.01

* Intake temperature on this date was 14°C but cultures were started at 20°C due to equipment failure.

Fig. 64. Growth rate ratio of *Oithona brevicornis*
as a function of discharge temperature.
For temperatures with more than one
observation, the vertical line represents
one standard error above and below the
mean value.

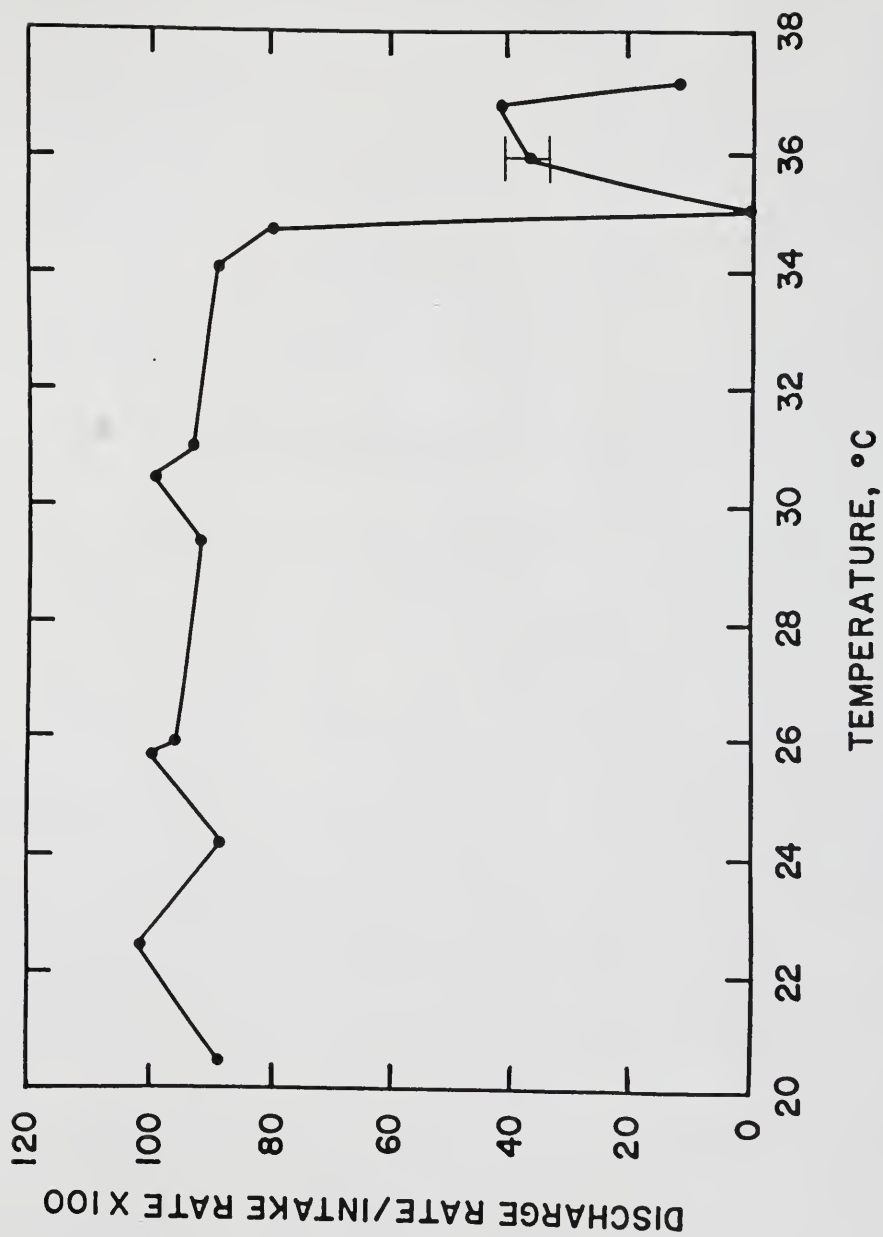
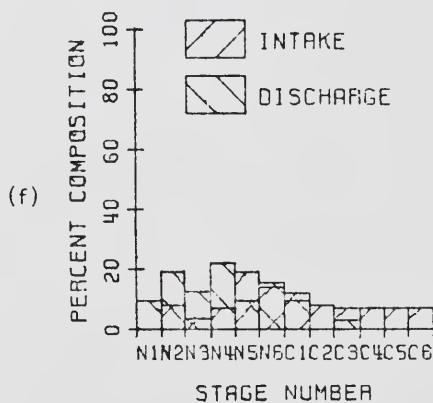
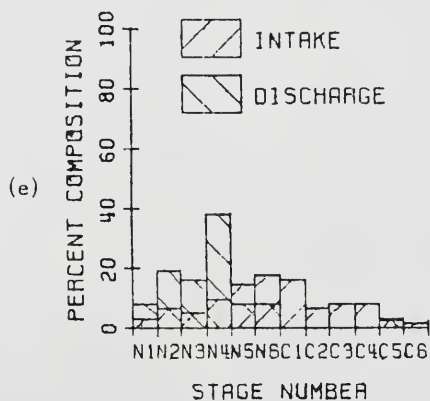
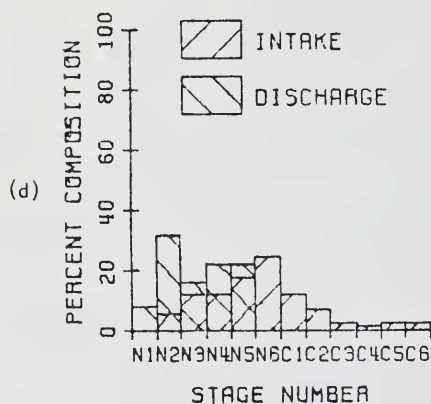
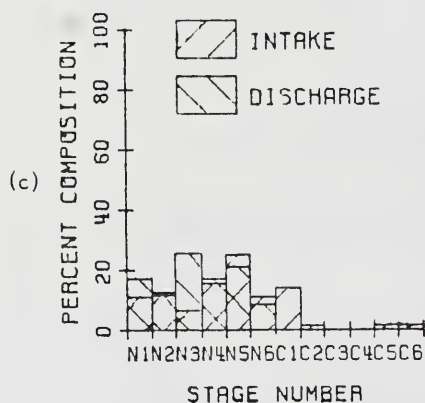
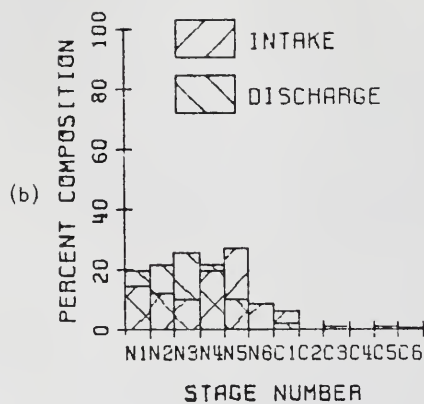
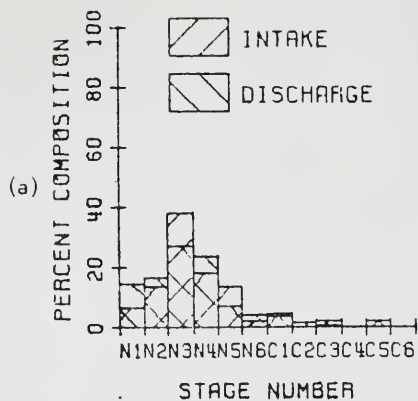


Fig. 65. Histogram series for Oithona brevicornis in growth experiment started on August 20, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 4, (e) Day 5, (f) Day 6. N1 to N6 are the six naupliar stages, while C1 to C5 are the five copepodite stages. C6 is the adult.



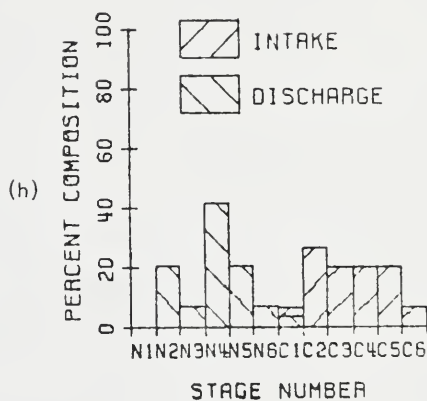
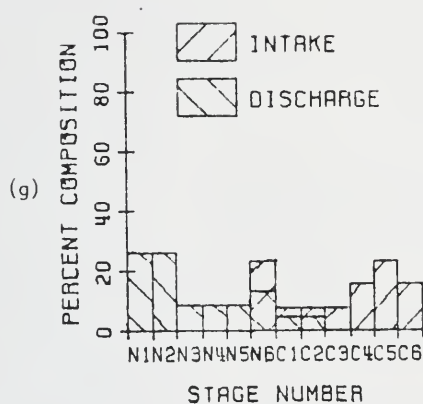


Fig. 65. Continued
(g) Day 7, (h) Day 8.

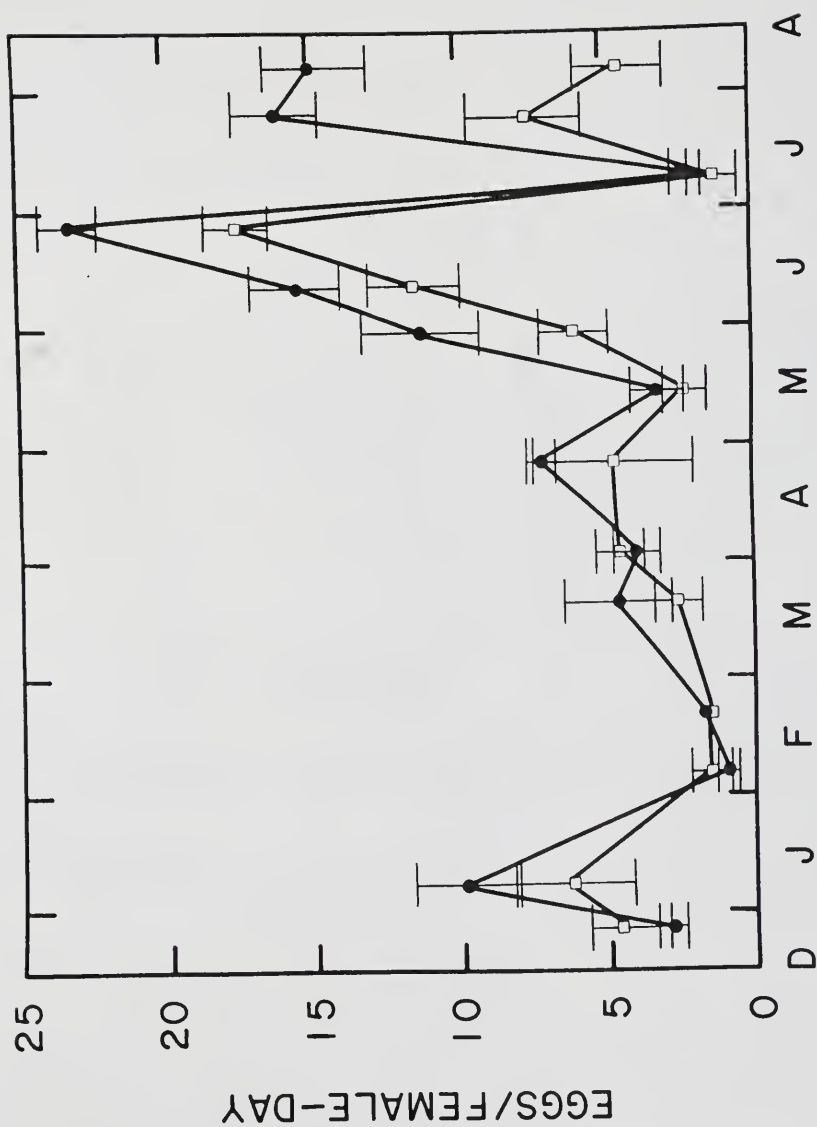
of 0. brevicornis juveniles began the experiment with a high percent of the population as early nauplii. As the days of the experiment proceeded the age structure shifted towards the older stages until, by day 8, all of the population were copepodites (primarily C4 to C6). The discharge population also started as early stage nauplii, but great shifts in age structure did not occur. By the end of the experiment, over 65% of the population were still nauplii. Only a small percentage of the discharge population ever achieved the older copepodite stages reached by the majority of the control population. The remainder of the population appeared to either grow at an extremely slow rate or not at all.

Reproductive Study of *Acartia tonsa*

Acartia tonsa adults reproduced well in laboratory culture, though the average eggs/female-day ratio varied from month to month (Figure 66). The trend in *Acartia tonsa* reproductive rates through the seasons traced that of naupliar densities (Figure 58) as well as numbers of older juveniles and adults (Figure 4). There were peaks in *Acartia tonsa* production in January, July and August. The peaks in reproductive rates appear to come two weeks ahead of the peaks in numbers, but the dates plotted are collection dates and the period for which the rates were measured ended one week later. Therefore, there appears to be a good agreement between trend in field measurements and reproduction rates found in the laboratory.

The reproductive rate of the intake *Acartia tonsa* ranged from 0.78 eggs/female-day to 23.2 eggs/female-day with a mean value of

Fig. 66. Seasonal variation in reproductive rate for intake (closed circles) and discharge (open squares) populations of Acartia tonsa. Vertical lines represent one standard error above and below mean values.



8.0 eggs/female-day. Discharge Acartia tonsa adults exhibited reproductive rates of 1.30 eggs/female-day to 17.5 eggs/female-day, with a mean value of 5.3 eggs/female-day. Multiple regression analysis showed that the sampling area from which the adults were collected (intake or discharge) and the date on which the samples were taken were both significantly related (0.001 level) to reproductive rates. Since it appeared that the reproductive rates of the intake and discharge populations were similar until the end of May, the year was divided into two parts and multiple regression analysis performed on each. Regression analysis for the experiments from December to May 14th showed no significant differences (above 0.05 level) in reproductive rates between the populations. There was, however, a highly significant difference (0.0002 level) attributable to entrainment for the experiments from the middle of May to the middle of August when experiments had to be discontinued because of the low survival of entrained adults.

To check further the significance of this period of reproductive depression, a Duncan's Multiple Range test from the SAS computer program package was run on data from the reproductive studies. Results from this test showed that reproductive rates for entrained Acartia tonsa were significantly lower (0.05 level) than those of control populations for the experiments performed during the period from the middle of May to the middle of August. The only exception was the reproductive study done on the 9th of July, when both intake and discharge populations exhibited low egg production. The mean reproductive rate of control populations of A. tonsa over the period of significant depression was 14.9 eggs/female-day while entrained populations averaged 41% lower at 8.8 eggs/female-day.

The relationship between discharge temperatures and entrainment effects on reproduction are shown in Figure 67. The ratio of discharge to intake reproductive rates fluctuated around the 100% level until temperatures rose above 34°C. Above this temperature, values for the reproductive rate ratio declined, with the lowest level being seen in August for a temperature of 35.1°C.

Long-Term Survival of *Acartia tonsa* Juveniles

Data from growth experiments were used to construct survivorship curves for intake and discharge populations of *Acartia tonsa* juveniles (Appendix C). Statistical comparison of mortality rates showed no significant differences (at 0.05 level) between the survival of the two populations for any of the experimental periods (Table 3). The mean mortality rate for intake populations of *A. tonsa* juveniles was 10.0%/day while that of discharge populations was 7.5%/day. Regression analysis of the data showed, however, that throughout the year there was no significant relationship (at 0.05 level) between juvenile mortality rates and whether or not initial populations had been entrained. Thus, the differences between the average mortality rates of the two populations was attributable to natural variation. Similar analysis showed that culturing temperature was also not a significant factor in determining mortality rates through the year.

Long-Term Survival of *Oithona brevicornis* Juveniles

Growth experiments on *Oithona brevicornis* also provided data for the analysis of long-term survival. Survivorship curves created from

Fig. 67. Reproductive rate ratio of Acartia tonsa
as a function of discharge temperature.
For temperatures with more than one
observation the vertical line represents
one standard error above and below the
mean value.

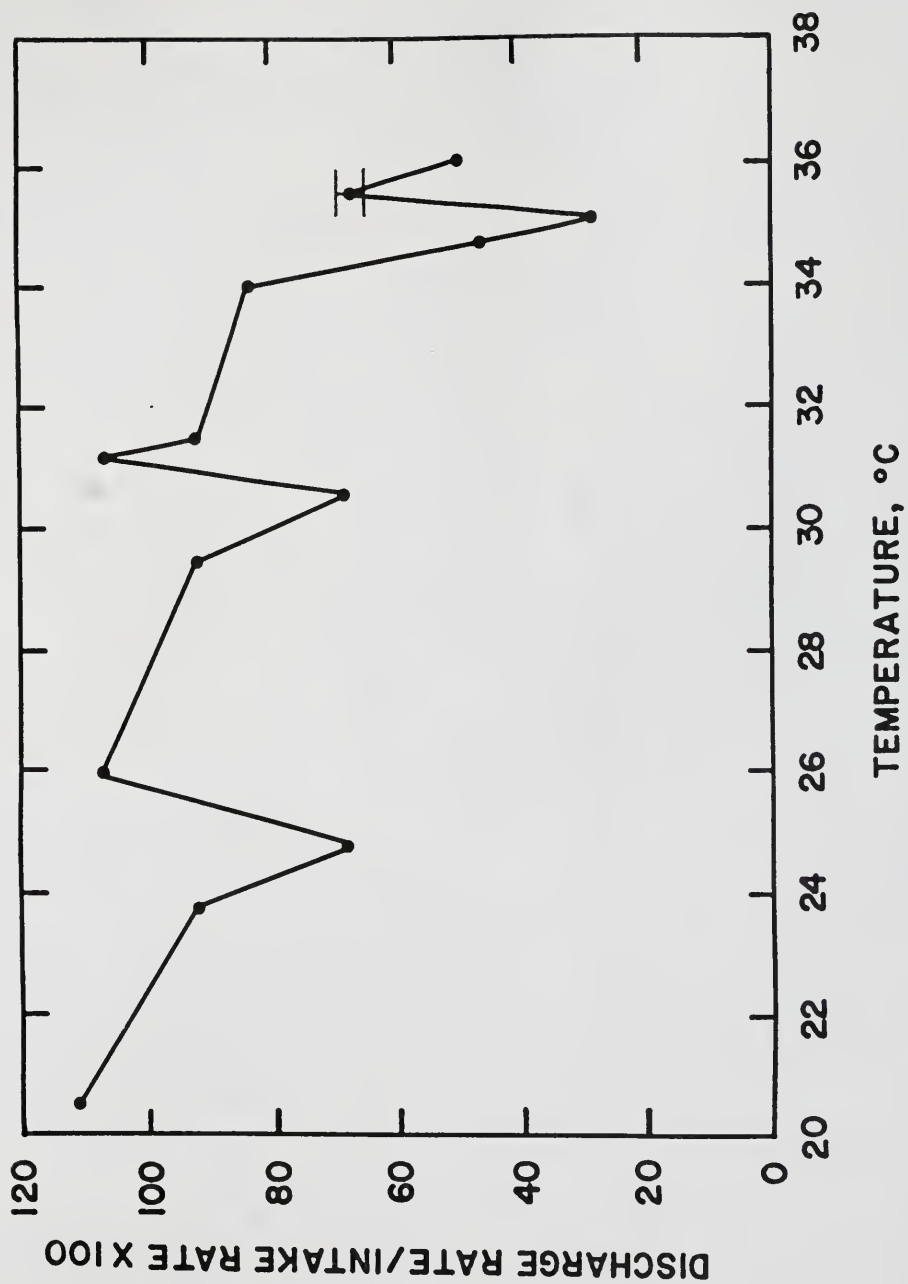


TABLE 3

Comparison of the Mortality Rates of Intake and Discharge *Acartia tonsa* Juveniles

DATE	TEMPERATURE (°C)		MORTALITY RATE (% per day)		T-VALUE	LEVEL OF SIGNIFICANT DIFFERENCE
	Intake	Discharge	Intake	Discharge		
11-13-73	19.0	24.7	7.5	13.0	0.29, 6df	N.S.
11-27-73	23.8	29.5	7.2	1.2	0.71, 8df	N.S.
12-26-73	20.0*	21.5	6.9	5.3	0.33, 10df	N.S.
01-22-74	20.4	24.3	4.6	5.5	0.12, 12df	N.S.
02-05-74	21.0	25.8	6.8	6.4	0.11, 12df	N.S.
03-19-74	20.2	26.0	9.9	4.8	0.36, 7df	N.S.
05-28-74	27.5	34.5	14.2	4.9	0.92, 6df	N.S.
06-11-74	29.5	36.0	31.5	8.2	0.84, 3df	N.S.
06-25-74	27.2	34.2	9.5	9.8	0.07, 12df	N.S.
07-09-74	28.8	35.7	13.6	8.9	0.87, 11df	N.S.
07-23-74	27.8	36.0	3.0	8.5	1.16, 12df	N.S.
08-06-74	28.9	35.1	14.1	8.2	0.97, 10df	N.S.
08-20-74	30.1	37.2	10.3	11.0	0.10, 10df	N.S.
09-03-74	30.9	37.0	7.1	8.2	0.17, 10df	N.S.

* Intake temperature on this date was 14°C but cultures were started at 20°C due to equipment failure.

these data are shown in Appendix C. Statistical comparison of the mortality rates of entrained and control populations of O. brevicornis juveniles showed no significant differences (at 0.05 level) for any of the experimental periods (Table 4). The mean mortality rate for intake juveniles was 8.4%/day, while that for discharge was 6.8%/day. The difference between the two rates was not significant (at 0.05 level), however, as regression analysis showed no significant relationship between entrainment and long-term survival as measured by post entrainment mortality rates. Culturing temperature was also shown not to be significant (at 0.05 level) in determining mortality rates of juvenile O. brevicornis through the year.

Long-Term Survival of Acartia tonsa adults

Observations from the pair-mating reproductive experiments allowed the creation of survivorship curves for Acartia tonsa adults (Appendix C). Statistical comparison of the mortality rates of intake and discharge populations obtained from these curves showed that there were no significant differences (at 0.05 level) in survival between the populations for any of the experimental periods (Table 5).

The total mortality values, as well as the average percent mortality/day, for intake and discharge populations in each reproductive experiment are shown in Table 6. The mortality rates calculated from the total mortality values had a mean value of 5.93%/day for intake Acartia tonsa and 6.47%/day for discharge populations. Regression analysis of the mortality rates showed, however, that there were no significant differences (at 0.05 level) between the populations. There

TABLE 4
Comparison of the mortality rates of Intake and Discharge *Oithona brevicornis* Juveniles

DATE	TEMPERATURE (°C)		MORTALITY RATE (% per day)		.T-VALUE	LEVEL OF SIGNIFICANT DIFFERENCE
	Intake	Discharge	Intake	Discharge		
11-27-73	23.8	29.5	4.8	2.6	0.48, 12df	N.S.
12-26-73	20.0*	21.5	9.9	7.4	0.33, 10df	N.S.
01-22-74	20.4	24.3	5.3	7.3	0.20, 12df	N.S.
02-05-74	21.0	25.8	13.8	13.0	0.05, 6df	N.S.
03-19-74	20.2	26.0	7.6	7.7	0.36, 7df	N.S.
04-24-74	23.9	30.6	7.8	6.6	0.16, 12df	N.S.
05-14-74	25.1	31.3	.0	2.6	1.40, 10df	N.S.
05-28-74	27.5	34.8	9.3	8.2	0.92, 6df	N.S.
06-11-74	29.5	36.0	22.0	13.9	0.41, 5df	N.S.
06-25-74	27.2	34.4	10.1	7.8	0.41, 12df	N.S.
07-09-74	28.8	35.7	.0	1.1	0.34, 12df	N.S.
07-23-74	27.8	36.0	6.9	3.6	0.61, 12df	N.S.
08-06-74	28.9	35.1	4.3	1.1	0.91, 12df	N.S.
08-20-74	30.1	37.2	12.1	9.5	0.43, 12df	N.S.
09-03-74	30.9	37.0	13.5	8.5	0.91, 10df	N.S.

* Intake temperature of this date was 14°C but cultures were started at 20°C due to equipment failure.

TABLE 5
Comparison of the Mortality Rates of Intake and Discharge Acartia tonsa adults

DATE	TEMPERATURE (°C)		MORTALITY RATE (% per day)		T-VALUE	LEVEL OF SIGNIFICANT DIFFERENCE
	<u>Intake</u>	<u>Discharge</u>	<u>Intake</u>	<u>Discharge</u>		
04-02-74	24.8	30.7	8.2	6.1	0.26, 10df	N.S.
05-14-74	25.1	31.3	8.6	6.3	0.17, 8df	N.S.
05-28-74	27.5	34.8	6.3	5.7	0.04, 8df	N.S.
06-11-74	29.5	36.0	3.1	7.9	0.36, 8df	N.S.
06-25-74	27.2	34.2	8.6	6.4	0.16, 8df	N.S.
07-09-74	28.8	35.7	19.2	19.6	0.02, 6df	N.S.
07-23-74	27.8	36.0	3.0	5.6	0.22, 8df	N.S.
08-06-74	28.9	35.1	10.9	14.2	0.20, 7df	N.S.

TABLE 6

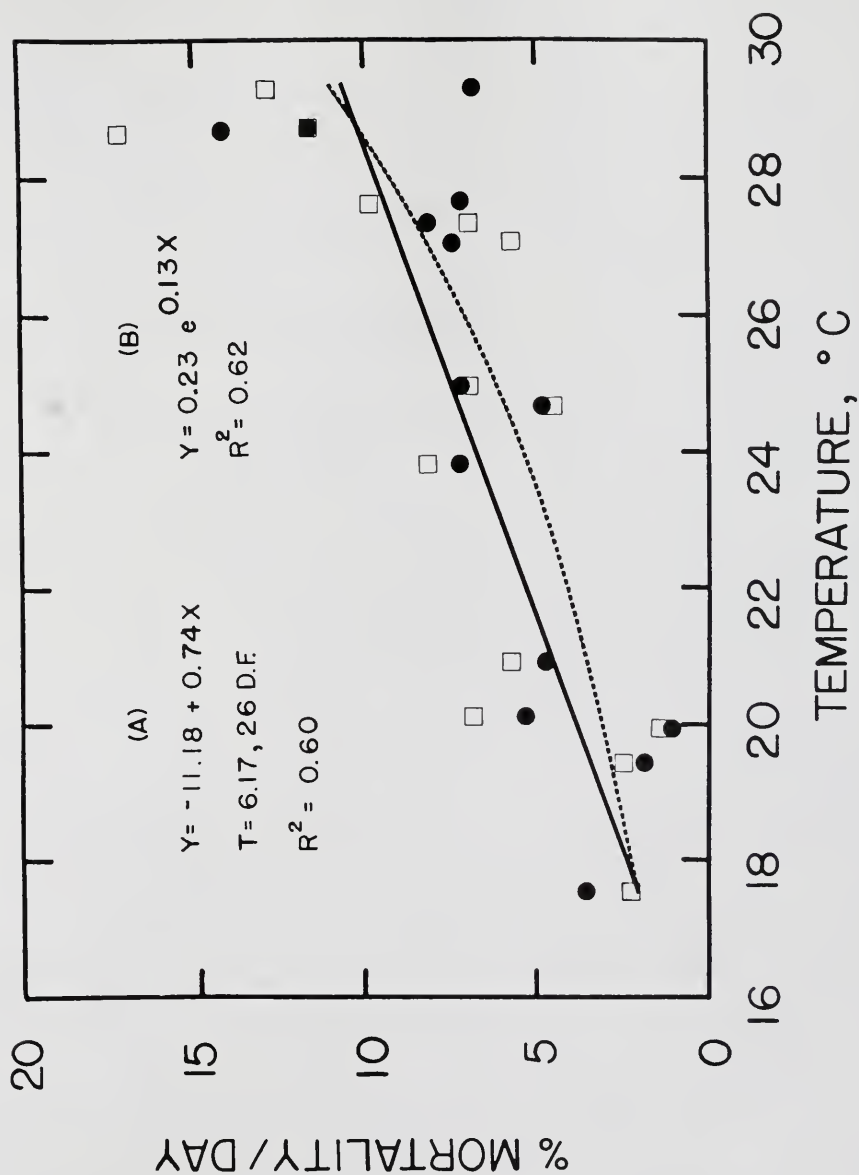
Average Percent Mortality/Day Calculated for Acartia tonsa Adults in Reproductive Experiments

DATE	TEMPERATURE	TOTAL MORTALITY OF EXPERIMENTAL PERIOD (%)		LENGTH OF PERIOD (days)	MORTALITY RATE (%/day)	
		Intake	Discharge		Intake	Discharge
12-26-73	20.0*	7.1	10.7	7	1.0	1.5
01-08-74	19.5	13.2	18.3	7	1.9	2.6
02-05-74	21.0	33.3	41.6	7	4.8	5.9
02-19-74	17.6	25.0	16.7	7	3.6	2.4
03-19-74	20.2	37.9	50.0	7	5.3	7.1
04-02-74	24.8	33.8	33.1	7	4.8	4.7
04-24-74	23.9	50.0	58.3	7	7.1	8.3
05-14-74	25.1	35.7	35.7	5	7.1	7.1
05-28-74	27.5	40.0	35.7	5	8.0	7.1
06-11-74	29.5	33.3	65.0	5	6.7	13.0
06-25-74	27.2	37.5	29.2	5	7.5	5.8
07-09-74	28.8	58.3	87.5	5	11.7	17.5
07-23-74	27.8	35.7	50.0	5	7.1	10.0
08-06-74	28.9	58.3	58.3	5	11.7	11.7

* Intake temperature of this date was 14°C but cultures were started at 20°C due to equipment failure.

was a highly significant (0.0005 level) relationship between mortality rate and culturing temperature (Figure 68), with higher temperature producing higher adult mortality rates. An exponential relationship fit the data slightly better than the linear equation, though neither provided a perfect fit. Natural variation in such factors as age of the adults and water characteristics at the time of collection would probably explain fluctuations within the trend.

Fig. 68. Mortality rate of Acartia tonsa adults as a function of culturing temperature. The closed circles are the intake population values, while the open squares are the discharge population values. The solid line is the linear regression fitted curve described by equation A while the broken line is the exponential fitted curve described by equation B.



SECTION IV

DISCUSSION

Field Studies

Copepods Species of the Crystal River Area

The species composition and relative abundance of copepods at Crystal River is similar to that found by investigators in other estuaries in Florida and the southeast (Davis, 1950; Fleminger, 1956; Woodmansee, 1958; Grice, 1957, 1960b; Hopkins, 1966; McIlwain, 1968; and Reeve, 1970). Greatest numbers of copepods appear in the summer and are for the most part dominated by a few species: Acartia tonsa, Oithona brevicornis and Paracalanus crassirostris. Moderate numbers of Euterpina acutifrons and Pseudodiaptomus coronatus, as well as sporadic occurrences of Labidocera spp. and Tortanus setacaudatus, made up the major portion of the remaining copepod species at Crystal River.

In terms of biomass, Acartia tonsa is generally considered the predominant estuarine copepod in Florida (Davis, 1950; Woodmansee, 1958; and Reeve, 1970). In the Crystal River area, this species reproduced year-round and exhibited highest numbers in early summer. Reeve (1970) has stated that A. tonsa appears not to be greatly restricted by variations in temperature or salinity conditions. This tolerance would account for the fact that this species represents a large portion of the copepod population through the year. Acartia tonsa comprises

the majority of the copepod population in winter and decreases in relative importance towards summer as other species gain in abundance.

Oithona spp. are numerically the dominant species at Crystal River. These species, primarily O. brevicornis, have been observed in great abundance in inshore areas (Deevey, 1956; Grice, 1960b, Hopkins, 1966; and Reeve, 1970). The small size of these species, however, prevented adequate sampling by investigators using large mesh nets (Woodmansee, 1958) and may have caused underestimates by the majority of workers. Even in the Crystal River study, some of the copepodite stages of these species were observed to pass through a 75 micron net. The loss of a portion of the juvenile population explains why the number of juveniles is at times relatively low compared to adult females (Figure 12). The large numbers of juveniles filtered into the initial growth experiment samples (Figure 62); and Appendix B, day 0.3 of each histogram series for O. brevicornis, also attests to the movement of some of the copepodite stages through the 75 micron net. The 158 micron nets used by most of the previous investigators would have certainly lost a sizable portion of the juveniles of these species.

Oithona spp. had a slight peak in numbers in December, but generally rose from lowest numbers in the winter to highest numbers in the summer. The small peak in December could represent the relative abundance of Oithona nana which has been observed to be a winter form (Woodmansee, 1958; and Reeve, 1970). The December samples were the only ones in which qualitative examination of the samples showed large numbers of female O. nana bearing egg-sacs. The trend in Oithona spp. abundance for the remainder of the year is probably due to domination by O. brevicornis. This species, though it breeds year-round, has

been shown to exhibit highest numbers during the warmest months (Deevey, 1956; Hopkins, 1966; and McIlwain, 1968). Oithona spp. comprise a majority of the total copepod population for most of the year but their small size probably make them less important than Acartia tonsa in contribution of biomass to the secondary production of the Crystal River area. Reeve (1970) has estimated that although Oithona contributed up to 80% of the total numbers of zooplankters of Biscayne Bay, Florida, it accounted for only 27% of the total biomass.

Paracalanus crassirostris was the third most abundant species in the Crystal River area. This species has been observed to be a significant member of coastal and estuarine zooplankton communities (Davis, 1950; Fleminger, 1956; Deevey, 1956; Grice, 1957, 1960b; and Hopkins, 1966). The small size of P. crassirostris may have caused many investigators to underestimate total numbers of this species since a portion of the juveniles and males of this species would pass through a 158 micron mesh plankton net. Grice (1957) has suggested that this species may have also been overlooked by investigators who considered it a developmental stage of Paracalanus parvus. Numbers of P. crassirostris at Crystal River rose from low levels in winter to highest levels in spring and summer. This species has also been observed to be a summer form in other estuaries (Deevey, 1956; and Hopkins, 1966), though Grice (1957) showed it to be abundant in coastal waters throughout the year.

Euterpina acutifrons is a small harpacticoid that was the fourth most abundant species at Crystal River. This species was found to be common in previous estuarine and coastal studies (Davis, 1950; Grice, 1957; Woodmansee, 1958; Hopkins, 1966; and McIlwain, 1968). Numbers

of E. acutifrons peaked in December, March and June. The general trend, however, was for numbers to decrease with the coming of summer, so that, by September, the species was almost absent from the collections. Woodmansee (1958) considers E. acutifrons a winter copepod in south Florida, while Grice (1957) observed no seasonal variation in numbers in north Florida. Hopkins (1966) and McIlwain (1968), also working in northern Gulf estuaries, observed this species to exhibit highest numbers in the spring and decrease in abundance through the summer. The Crystal River area may represent an intermediate point in geography and temperature regime between the south and north Florida estuaries, so that this species is relatively abundant through the winter and spring, but almost disappears during the hottest months. Temperature is probably not the only factor affecting abundance of this species, however, because environmental factors such as salinity, phytoplankton production, competition with other zooplankters and current patterns may all affect seasonal distribution.

Pseudodiaptomus coronatus has been observed to be an estuarine species that seasonally occurs in significant numbers (Deevey, 1956; Grice, 1957; Woodmansee, 1958; and Hopkins, 1966). At Crystal River, this species was relatively abundant from December through March, but decreased in importance in the spring and summer. Woodmansee (1958) considers P. coronatus a fall-winter copepod in south Florida and his review of previous literature shows the seasonal trend to hold true generally for most areas. This species does not, however, appear to be restricted by temperature (Woodmansee, 1958) or salinity (Deevey, 1956), so that seasonal abundance appears to be controlled by other environmental factors. One possible factor is that the adult

P. coronatus is largely epibenthic (Jacobs, 1961) and appears to feed, at least in part, on attached benthic microalgae and epiphytes (personal observation from laboratory culture). The production of these benthic algae would tend to lose dominance to the phytoplankton blooms that are observed to occur in the spring and summer at Crystal River. The drop in numbers of P. coronatus may, therefore, reflect the seasonal shifting of food source to other species that are more fully pelagic.

The partially epibenthic lifestyle of P. coronatus adults also explains the fact that relatively few adults were collected, even in samples in which juveniles were abundant (Figure 34). It has also been noted (Deevey, 1956; and Maturo et al., 1974) that P. coronatus adults are found sporadically in large numbers during night collections. These observations could be attributed to a natural diurnal migration of this species from the bottom at night or may be a function of breeding activities, as males appear to seek active females in the water column (Jacobs, 1961). Whatever the cause, these observations indicate that the importance of this species to the secondary production of an area may be underestimated by counts from daytime plankton tows.

Labidocera spp. found at Crystal River consist of Labidocera aestiva, a northern species, and Labidocera scotti, a tropical species. Fleminger (1956) has discussed in detail the distribution of these species in the Gulf of Mexico. These species were observed in some number in winter, but the major period of abundance was in the summer. Although the species were not separated due to the difficulty of copepodite identification (Woodmansee, 1958; and Reeve, 1970),

qualitative observations of the samples indicated that the winter peak represented relatively higher numbers of L. scotti and L. aestiva dominated the summer period of abundance. Woodmansee (1958) and Reeve (1970) have observed that L. scotti breeds in south Florida waters during the winter. It appears that the Florida west coast counter current that distributes L. scotti along the coast (Grice, 1957) brings relatively high numbers of this species to the Crystal River area during the winter breeding season. The relative abundance of this species may, therefore, reflect high numbers in tropical source waters, rather than an actively breeding population. Grice (1957) stated that no spermatophore-bearing females were observed in Gulf waters. This observation plus the fact that there are relatively low numbers of juveniles compared to adults in the Labidocera spp. category during winter (Figure 40), suggests that L. scotti does not reproduce well in the colder waters of the Crystal River area. Labidocera aestiva, on the other hand, were qualitatively observed to dominate the summer counts of this category and appeared to reproduce well. A large peak of Labidocera spp. juveniles occurred in July and numbers were relatively high during the entire summer. Hopkins (1966) and McIlwain (1968) reported that L. aestiva reached highest numbers during these warmer months in estuaries in the northern Gulf of Mexico. It appears that, although the Crystal River estuary receives some influence from the tropical species L. scotti, the temperate species L. aestiva is more important to the zooplankton production of the area.

The large size of Labidocera spp. prevents the estimation of importance of these species by numbers alone. Reeve (1970) has observed that, although Labidocera spp. annually represented only 1%

of the total zooplankton population of Biscayne Bay, these species represented 15% of the total biomass. Using the same conversion ratio of 15 to 1, Labidocera spp. may at times represent up to 40% of the biomass of the copepod population at Crystal River. These species may therefore seasonally represent important producers in the Crystal River estuary. Equally important to the ecology of the area is the fact that at least part of the biomass of the Labidocera spp. is produced by predation on other copepod species (Fleminger, 1956).

Tortanus setacaudatus has been observed in moderate numbers in coastal and estuarine waters of the Gulf of Mexico (Grice, 1957); and Hopkins, 1966). These observations have shown that this species occurred in significant numbers only during the warmer months. The abundance of this species at Crystal River followed this trend, with peak numbers occurring in April and during the late summer. Perhaps the primary significance of this sporadically occurring species is that it has been observed to be a voracious predator. As many as three late stage Acartia tonsa copepodites have been observed to be captured and partially consumed by a female T. setacaudatus in the course of an hour (personal observation from laboratory culture). Of course, such observations exaggerate estimates of capture rate because of the restriction of escape behavior by prey under laboratory conditions, but it appears that this species could be an important copepod predator when in abundance in the estuary.

Factors Involved in Entrainment Mortality

Temperature and salinity effects

The response of the various copepods to the effects of entrainment show certain similarities, as well as individual differences. Temperature, salinity, and temperature-salinity interactions are the most important factors in determining the resistance of the copepods to thermal stress (ΔT of approximately 6°C). All of the species appear to have a critical thermal level above which mortality increases exponentially. Below this level, however, the response pattern for each species has a unique configuration which is shaped by the ecological requirements and biological characteristics of the species.

The response pattern of Acartia tonsa displays an area of resistance to thermal shock for quite a wide range of temperatures under conditions of low salinities. This species is euryhaline but high salinities (above 25 ppt.) are considered suboptimal (Barlow, 1955; and Riley, 1967). Gonzalez (1974) reports that Acartia tonsa populations taken from various estuarine areas are eurythermal, but makes no mention of salinity conditions. The response surface for A. tonsa seems to reflect these observations, with lower salinities allowing complete survival up to 35°C . Higher salinities, which are probably suboptimal, produce greater mortalities when the species is subjected to thermal shock. A slight high salinity-high temperature tolerance interaction is observed for A. tonsa populations under thermal stress. The beneficial effects of such high/high combinations on poikilotherms have been described by Ranarde (1957), Lance (1963), Alderdice (1972) and most extensively by Kinne (1964).

For temperatures below 35°C, Oithona spp. show quite low mortalities when exposed to short periods of thermal shock. Mortality is linearly related to temperature except that, as with Acartia tonsa, high salinities appear to buffer the effects of moderately high temperature. Other than this interaction, salinity plays little part in the resistance of Oithona spp. to thermal shock. Oithona spp., principally O. brevicornis, are estuarine species which are abundant in most areas with salinities below 31 ppt. (Grice, 1957). Apparently, since the salinities observed during the study fall within the optimum range for Oithona spp., temperature is the major lethal factor.

Paracalanus crassirostris is a coastal species, generally found in areas with salinities of 24 to 38 ppt. and temperatures of 10 to 32°C (Grice, 1960b). This species demonstrates quite a resistance to the immediate effects of thermal shock. There is a temperature-salinity interaction exhibited by the response pattern of this species, with high salinities being associated with low mortalities across the range of temperatures. No combination of these factors, however, produces mortalities above 70%.

Euterpina acutifrons was shown by Grice (1957) to have an identical distributional range with Paracalanus crassirostris. It is not surprising, therefore, that this species shows a temperature-salinity interaction of the type described for P. crassirostris. Salinity, however, seems to play a more important role in the amount of mortality caused by thermal shock. High mortalities are correlated with low salinities, especially in combination with high temperatures.

Since multiple regression analysis could not be performed for Pseudodiaptomus coronatus, Labidocera spp. or Tortanus setacaudatus,

only general qualitative observations can be made about the response of these species to thermal shock. All three species exhibited low to moderate mortalities through the year except for T. setacaudatus, which showed 100% mortality for the hottest sampling data in August. In general, it appears that these species are quite resistant to the immediate effect of thermal shock for all but the very highest temperatures.

Delayed thermal effects

The effects of entrainment on the survival of zooplankters involve not only the ambient temperature and the amount of heat being added to the water (ΔT), but the duration of exposure to the elevated thermal conditions as well (Coutant, 1972, 1974; and Davies and Jensen, 1974). This premise has been confirmed by recent investigators (Icanberry and Adams, 1974; Lauer et al., 1974; and Prager, 1971) who have shown that significant increases in zooplankton mortality are related to increased duration of exposure to the thermal effluent. In the Crystal River study, the effects of exposure for the two hours that the entrained population remains in the discharge canal produce observable changes in the response patterns of the various species.

The high salinity-high temperature interaction exhibited by Acartia tonsa starts to disappear when the species is exposed to thermal stress for a longer period of time. Temperature increases above the 35°C level produce a greater increase in mortality when this species remains in the heated effluent. The response patterns of Oithona spp. exhibit similar effects, with the high salinity-high

temperature interaction being reduced in importance and mortalities for temperatures above 35°C increasing rapidly to the 100% level.

The responses of Paracalanus crassirostris and Euterpina acutifrons to several hours of thermal stress were similar. The initial resistance to thermal shock disappears and both species show a sensitivity to high temperatures (above 33°C) for all salinities. The temperature-salinity interaction remains important for temperatures below 33°C and the optimum conditions for resistance to thermal stress (within the realm of this study) center around the combination of 25°C and 30 ppt. Males of both species were particularly sensitive to extended exposure to elevated temperatures, showing mortalities 10 to 15% higher than females.

The effects of extended thermal stress seem to be less important for the estuarine species Oithona spp. and Acartia tonsa than for the coastal species Paracalanus crassirostris and Euterpina acutifrons. The estuarine species exhibit low mortalities over a wide range of temperature and salinity combinations and show little increase in mortality due to canal effects except at highest temperatures (above 35°C). The coastal species, on the other hand, have low mortalities only in areas of high salinity and low temperatures. The canal effects cause temperatures much above 33°C to become lethal for these species. To use terms defined by Alderdice (1972), Oithona spp. and Acartia tonsa are euryplastic with respect to resistance to thermal stress, while Paracalanus crassirostris and Euterpina acutifrons are stenoplastic. These observations hold with the generally accepted maxim that estuarine species are better adapted to

resist large fluctuations in temperature and salinity conditions than are either coastal or oceanic forms.

The effect of the two-hour exposure to elevated temperatures on the less common species of the Crystal River area can only be discussed in general qualitative terms. Pseudodiaptomus coronatus appeared to be virtually unaffected by the prolonged exposure to the heated effluents until mid-summer. This seems to indicate that P. coronatus is fairly resistant to exposure to thermal stress despite the fact that it appears to be a winter species. Labidocera spp. appeared to be much affected by the increased duration of exposure to the elevated temperatures. When subjected to treatment ID2, these species showed high mortalities for most of the summer sampling dates. Since Labidocera aestiva made up the major portion of the Labidocera spp. category for this season, it appears that this temperate species is living close to its thermal limits in the Crystal River area during the summer and is sensitive to prolonged exposure to thermal stress at this time. Fleminger (1956) placed the southern limit of the biogeographic range of this species at the central portion of the west coast of Florida, which indicates that L. aestiva found in the Crystal River area are probably living under suboptimal conditions of natural thermal stress. The response of Tortanus setacaudatus to the prolonged exposure to heat within the discharge canal appeared to show sensitivity only to the highest temperatures in summer. Prager (1971) described this species as being a tidepool and mangrove swamp dweller which appears to have adapted to thermal stress. The observations of T. setacaudatus at Crystal River confirm the hardiness of this species, which is especially obvious in the comparison of the

mortalities shown in Figure 10g to those of other copepod species that are shown in Figure 10. It is only at the highest temperatures (above 36°C) that mortalities of T. setacaudatus rise exponentially, whereas species such as Paracalanus crassirostris and Euterpina acutifrons exhibit mortality increases at much lower temperatures.

Mechanical effects

The major effect of entrainment other than thermal stress is the mechanical damage. Such physical damage is caused by the shearing forces and turbulences encountered in passage through great lengths of small diameter pipes of the condenser system of the power plant. Differences in plant design and capacity make the mechanical effects of entrainment on zooplankters unique for each site, so that statements made for one location cannot be generalized to apply to all others. Mechanical-thermal interactions confound the effects of the mechanical damage of entrainment because zooplankters coming in contact with the hot walls of condenser pipes are exposed to hotter conditions than are indicated by discharge water temperatures. The effects of this interaction also vary among sites because of differences in factors such as pumping capacity and speed, numbers and size of condenser pipes, efficiency of the cooling system and electrical output of the power plant. Marcy (1974) has given an extensive review of entrainment studies that have looked at the mechanical effects of entrainment. He reports that such studies have shown entrainment mortality caused by mechanical damage ranged from negligible levels to nearly 100%, depending upon the particular

species of zooplankter and power plant being observed. The general consensus of all the reports was that the mechanical damage of entrainment depended upon the size and susceptibility of the species to physical stress as well as the design of the power plant.

The mortality caused by mechanical damage associated with passage through the Crystal River power plant seems to depend, at least in part, on the size of the entrained organism. Labidocera spp., by far the largest species of the area, exhibit high mortalities, regardless of environmental conditions (Figures 10e, 44 and 45). On the other hand, Oithona spp., the smallest copepods in the area, exhibit no significant mechanical damage. The remaining species, which are intermediate in size, undergo some mechanical damage due to entrainment.

Mechanical damage is correlated with a rise in mortality for Acartia tonsa females and juveniles of 3 to 5% for all but optimum conditions of temperature and salinity. The males of this species appear to be more sensitive and exhibit a rise of 10% mortality. Paracalanus crassirostris and Euterpina acutifrons also show a 5 to 10% rise in mortalities for low temperatures, but a mechanical-temperature interaction causes mechanical damage to be more lethal at higher temperatures. This interaction is particularly obvious with Euterpina acutifrons where temperature becomes the major lethal factor if the organisms have passed through the power plant. Such an interaction may be due to the fact that the species are sensitive to contact with the heated walls of the condenser pipes or may result from the thermally stressed organisms being more susceptible to the lethal effects of mechanical damage.

Although response surfaces were not made for Pseudodiaptomus coronatus and Tortanus setacaudatus, certain observations can be made from the data about the mechanical effects of entrainment on these species. Differences in mean mortality values between treatments that involved physical entrainment (DS0 and DS2) and those that involved thermal shock alone (ID0 and ID2) indicate that P. coronatus suffer mortalities 12 to 14% higher due to mechanical damage. The trend of mortalities through the year for treatment DS0 and DS2 roughly follows those for ID0 and ID2, indicating that no great mechanical-thermal interaction is observable. The size of the males and older juveniles of this species is roughly comparable to Acartia tonsa, while the female is quite a bit larger. It is, therefore, not surprising that P. coronatus show slightly higher mortalities due to mechanical damage than do A. tonsa.

Tortanus setacaudatus populations subjected to physical passage through the power plant show no differences in average mortality from those that were subjected to the same exposure to thermal shock alone (DS0 vs. ID0 and DS2 vs. ID2). This would indicate that there is little or no mechanical damage and that trends in mortalities are attributable to thermal effects. The trend is especially obvious in Figure 10g which shows entrainment mortalities are nearly 0% for all but the highest temperatures. Tortanus setacaudatus is slightly longer than Acartia tonsa, but is visually much more robust, having a heavily built body and thickened mouth parts. The difference in morphology may account for the resistance to mechanical damage by this species. The fact that T. setacaudatus is a tide-pool dweller (Prager, 1971) may also explain why this species apparently has adaptations to resist

the effects of turbulence, since surf action would be a periodic stress in such an environment.

Seasonal effects

Paracalanus crassirostris and Euterpina acutifrons both show lower mortalities than expected for spring season, while Acartia tonsa shows higher mortalities. The factors that have caused such responses are unknown, but may be related to phytoplankton blooms or changes in organic and inorganic constituents of the water associated with spring rains. Such factors may affect the overall health of the populations, which would in turn determine their response to entrainment effects. Bary (1964) has discussed how unknown characteristic of sea water affect biogeographical distribution of plankton which cannot be explained by temperature and salinity conditions. If the overall survival of a species in an area can depend on unknown constituents of the water, the seasonal changes in water characteristics could certainly affect the overall resistance of certain species to thermal stress.

Density effects

Paracalanus crassirostris and Euterpina acutifrons show lower entrainment mortalities when populations are high. The density variable may simply be an indirect measurement of the factors determining the general health of the population. High numbers would be found during optimum conditions and lower entrainment mortalities would be expected during this time when populations are healthiest.

Metabolic factors such as hormone levels may also have an effect on response patterns during breeding seasons.

Age and sex effects

Generally, juveniles of a given species show lower entrainment mortalities than the adults. For Oithona spp. and Paracalanus crassirostris, the difference in response between age classes was not significant, but Acartia tonsa and Euterpina acutifrons juveniles have significantly lower mortalities than the adults. Jensen et al. (1969) suggest that juvenile poikilotherms may not be as susceptible to thermal death as adults of the same species because they are biochemically less complex. The smaller size of the juveniles may also make them less likely to suffer physical damage in passage through the plant.

Males of a species are usually more sensitive to entrainment effects than the females. Even when the males were more resistant to the initial thermal shock, as in the case of Paracalanus crassirostris and Euterpina acutifrons, they were generally more sensitive to mechanical and canal effects, so entrainment mortality was higher. Hormonal and metabolic differences between males and females may produce differences in resistance to entrainment effects. Woodmansee (1958) explains the low male to female sex ratio found in natural populations of Acartia tonsa by suggesting that the males are more susceptible to natural mortality. Sex ratios for all major species studied show similar trends, suggesting that males of a species could

be the most sensitive to stressful conditions, such as those encountered in entrainment.

The less common species Pseudodiaptomus coronatus, Labidocera spp. and Tortanus setacaudatus did not provide enough observations for complete analysis of sex and age effects, but the general trends appear to hold true for most of the data available. Pseudodiaptomus coronatus and Labidocera spp. adult forms were found to be more sensitive to entrainment mortality than juveniles. Males of the Labidocera spp. appear to be slightly more sensitive than the females or juveniles, exhibiting close to 100% entrainment mortality throughout the year. The data from Tortanus setacaudatus showed considerable variation of response with respect to sex and age class. Juveniles appear slightly more sensitive to thermal stress, while adults appear more sensitive to the thermal-mechanical combination encountered in entrainment. Male T. setacaudatus, although quite rare, appear slightly more resistant to entrainment mortality compared to females, possibly because of their much smaller size.

Effects on the total copepod population

To give an overview of entrainment effects and summarize the significant factors involved, general trends in entrainment mortality of the entire copepod population can be observed.

Temperature is obviously the most important factor in determining the response of the copepods to entrainment. Mortalities are generally low for temperatures below 35°C and rise rapidly with small increases in temperature above this range. A temperature-salinity interaction appears evident, with mortalities being lower with

conditions of high salinities and moderately high temperatures and also with low salinities and low temperatures. The extended exposure of the copepods to the heated effluent tends to raise mortalities, especially at times of highest temperatures (above 35°C).

The mechanical effects of entrainment account for a moderate rise (10 to 20%) in copepod mortality. Although the mortality due to mechanical damage is relatively low, it appears to be the major lethal factor for all but the hottest months.

Male copepods exhibit higher entrainment mortalities than females or juveniles. This trend is more pronounced at higher temperatures, indicating that males are generally more sensitive to temperature-related aspects of entrainment.

Unknown seasonal factors cause entrainment mortalities of copepods in spring to be approximately 6% higher than can be explained by other conditions. This trend, as many of the others, reflects the dominance of Acartia tonsa, which is the species that shows sensitivity to springtime conditions.

Biological and Ecological Implications

Temperature has been shown to play the major role in determining the amount of entrainment mortality exhibited by copepods. A general trend seems apparent for the entrainment mortalities exhibited by the various species with respect to temperature (Figure 10). Mortalities fluctuate at moderately low levels until temperatures pass a threshold level, then rise in an exponential manner. This type of dramatic rise in

mortalities indicates that death has resulted from a temperature-induced enzyme deactivation that is most simply described by the Arrhenius equation (Johnson et al., 1974; and Eyring and Eyring, 1967):

$$V = Ae^{\frac{-E}{RT}}$$

where V represents velocity (in this case, mortality rate); A and R are constants; T is the absolute temperature; and E is the energy of activation. When temperatures rise above a critical threshold, there is enough energy of activation (E) to cause enzymes to be denatured at a faster rate than they can be replaced by the copepod and thermal death results. As the temperatures go above the critical point, the rate of denaturation shows an exponential increase and mortality values found for the two-hour period following entrainment also rise exponentially. Johnson et al. (1974) and Eyring and Eyring (1967) have examined aspects and implications of the Arrhenius theory and enzyme denaturation. Odum (1973) has proposed an analog simulation model of the effects of temperature on general biological systems with respect to the Arrhenius relation.

There has been considerable debate over which biological components are denatured in heat death. Bowler et al. (1973) give a review of the various schools of thought and conclude that there are two basic hypotheses on the cause of thermal death: denaturation of proteins, and the "melting" of cellular lipids. These investigators then propose a third possibility: heat injury results from the disruption of the stability of lipoprotein complexes in membranes and the

inactivation of enzymes (ATPase) that depend on their integrity. Each of these possible mechanisms have had supportive evidence and can explain the Arrhenius relationship. Quite possibly the same cause is not responsible for thermal death in different organisms, because heat injury is apparently complex and the most sensitive inactivation reaction may vary between biological systems.

The high salinity-high temperatures' resistance interaction seen for most species can be explained in terms of the various possible mechanisms of thermal death. Jensen et al. (1969) speculate that since the rate of denaturing of soluble protein complexes decreases with increasing initial concentration, high salinities would increase the water flow out of the organism and cause a beneficial concentration of the proteins. The osmotic permeability increases with temperature so this effect would be more pronounced as the temperatures rise. The salinity interaction might also be explained in terms of the hypothesis of Bowler et al. (1973) which states that heat injury of cells is primarily due to disruption of the sodium-potassium balance in membranes resulting from ATPase deactivation. Since ATPase activity is dependent on certain cations (Bowler and Duncan, 1967), one might speculate that increased salinities make more of these ions available within an organism and somehow slow the inactivation of the enzyme. The increased concentration of ions initially in balance across membranes may also affect the rate of disruption of the equilibrium. A third possible explanation of the temperature-salinity interaction is that most estuarine species must develop efficient osmoregulatory mechanisms to survive the fluctuating salinities of their environment (Jeffries, 1962). Such adaptations require energy that might

otherwise be used in the replacement of denatured substances and in the removal of the toxic by-products. High salinities would provide relatively less stress on these regulatory mechanisms and more energy could be used in maintenance of the substances that would otherwise be inactivated. Evidence of the stress of low salinities is shown by the increase in respiration observed when copepods are exposed to diluted sea water (Lance, 1965).

There are several ecological implications that may be drawn from the field study on entrainment. First of all, since high temperatures generally cause the major portion of the entrainment mortality of the copepods it can be assumed that the effects extend throughout the thermally impacted area. Copepods tidally recruited into the thermal plume of the power plant may exhibit nearly the same level of mortality as the copepods actually being entrained. The overall impact of the power plant during the summer months, therefore, affects much more of the copepod population than those actually passing through the condenser systems. The scope of additional temperature effects within the areas of the estuary influenced by the thermal effluent must be taken into account when the impact of the plant on the copepod standing crop and production is assessed.

Subtle effects of entrainment mortality involve changes in community structure, productivity and energy flow of the copepods population. The effects of such changes are not known because subtleties of relationships within food chains are not well understood. Natural variability in zooplankton populations make the changes difficult to observe, as it may take years for fluctuations in numbers to reach a point where the subtle selection forces of entrainment

become significantly important and produce alterations in community structure that can be seen.

An entrainment-induced trend that might be seen in the Crystal River area is a general reduction in size of individuals of the copepod population. The mechanical effects of entrainment cause differential mortality between large forms such as Labidocera spp. and small forms such as Oithona spp. This trend may have a double effect since the smaller species are freed from some of the predation of Labidocera spp. and therefore can increase in relative abundance all the more. Oithona spp. exhibit the least sensitivity to overall entrainment effects and would therefore be expected to gain competitive advantage in the ecosystem where the power plant acts as an artificial selection pressure. Even within a given species, differential mortality between age classes would tend to cause the more resistant juveniles to come into relative abundance compared to the larger adult forms. This general trend toward smaller organisms could have important implications to energy flow patterns of the estuary. Heinle (1969) has recognized the possibility of such a trend in the Patuxent estuary and has proposed that energy might be diverted from selective planktivores to passive filter feeders. The selective planktivores feed on certain size classes of copepods and include commercially important species such as anchovies and the juvenile forms of most estuarine fishes. The passive filter feeders, on the other hand, feed on all sizes of copepods and are mainly represented by forms such as ctenophores and jelly fish that form more or less a "dead end" in the food web. The shifting of energy to passive filter feeders might not only have direct adverse effects on the abundance of

commercially important planktivores but also may have effects on higher trophic levels that feed on planktivorous species. Such a shift could therefore eventually affect the pattern of energy flow for all organisms of the food web of an impacted area. The potential for such changes in patterns of energy flow is seen to be particularly great in estuaries where numbers of species are low and the options for a planktivore in the food web are few.

A second trend that might occur in the Crystal River area as a result of entrainment effects is a shift of energy from pelagic to benthic organisms. Heinle (1969) has reported that O. brevicornis is at times epibenthic and speculates that its resistance to thermal stress may divert energy to the benthos and away from less tolerant pelagic species. Trends in differential mortality between species at Crystal River indicate that Oithona spp. and Pseudodiaptomus coronatus are more resistant to entrainment effects than some of the other species. Since these species appear to be epibenthic at times, their tolerance of thermal stress could cause changes in community structure that would divert energy to the benthos that would normally go to predators of pelagic forms. The resulting shift in energy flow could have as far-reaching effects on the structure of the food web of the impacted area as those discussed for the trend of decreased size of the copepods.

Possibly the most obvious change in energy flow caused by entrainment is the diversion of a portion of the copepod population from the consumer food web to the decomposers. The copepods killed by entrainment effects quickly settle out of the water column and are either completely decomposed by bacteria and microbes or incorporated into

the food web of benthic detritivores. It is ironic that copepods, the organisms that are affected by entrainment, are generally considered the major link that transfers energy from the decomposers to higher trophic levels in the water column. Copepods appear to feed upon detrital particles suspended in the water and utilize the protein of the decomposers associated with them. The combined effect of greater input of dead organisms and the decreased number of living copepods to recycle the biomass of decomposers means that the benthic community is getting relatively more of the energy of the system and the nekton is receiving less.

The increased amounts of dead organic material made available to the decomposers could also mean that more nutrients will be released to the waters of the estuary. High nutrient levels could result in the same type of eutrophication as found in ecosystems with organic additions such as sewage (Weiss and Wilkes, 1974) or seafood wastes (Heald and Odum, 1974). In discussing entrainment mortality at a power plant on Biscayne Bay, Florida, Prager (1971) speculated that the dead zooplankters released back into the ecosystem might even be of legal concern as organic refuse under a waste disposal law. The changes that may occur in the estuary due to the increased organic input, as well as the size of the area influenced, depend on the amount of organic materials and the associated nutrients that can be naturally utilized by the system. Heald and Odum (1974) discuss the various effects of eutrophication found in estuaries with high organic loads, including blooms of small, atypical phytoplankters; patchy or stratified water with low oxygen content; reduced species diversity in the zooplankton and higher trophic levels; and altered community

structure and energy flow compared to similar, unpolluted systems. If this type of eutrophication were to occur due to entrainment-induced mortality of large numbers of zooplankters, the elevated temperatures of the impacted area may compound the problem by increasing the respiration and turnover rates of the organisms, while at the same time decreasing the oxygen carrying capacity of the water. Drastic ecological changes could occur as pollution-adapted organisms replace components of the natural ecosystem that cannot survive the eutrophic conditions.

The indirect ecological effects of entrainment and thermal pollution could have a spectrum of magnitudes. Some of the trends might be so subtle as to be negligible, while others may produce major changes within the ecosystem even if they are not readily apparent upon first observation. The very fact that changes in community structure, productivity and energy flow of the copepod population could occur due to the effects of entrainment suggests that such changes should be studied and effects predicted before the final assessment of environmental damage is made.

Finally, several implications on power plant design and siting can be drawn from the entrainment field study. Reeve and Cosper (1974) have speculated that tropical organisms live near their critical thermal limits during the warmest months of the year. This observation appears to hold true for copepods at Crystal River. Even with average ΔT values of less than 6°C , all of the species reached their thermal limits at some point during the summer. Power plant designers should keep this phenomenon in mind when designing cooling systems for power plants in tropical and subtropical areas.

A condenser system that provides the flexibility to adjust ΔT values seasonally would be most environmentally sound in southern areas, since the entrained organisms appear to be able to tolerate higher ΔT values in winter than in summer.

The duration of exposure to the thermal discharge plays an important role in the mortality of most species of copepods, so it can be inferred that returning the populations back to ambient temperatures as quickly as possible might greatly reduce overall entrainment effects. The elimination of long discharge canals which retain high discharge temperatures might help alleviate the problem. Discharge canals are designed to allow dissipation of excess heat before the effluent of the power plant returns to the estuary. At the Crystal River power plant, and probably at most subtropical and warm temperate sites, the process of heat dissipation is quite inefficient due to low surface area and high air temperatures. The discharge waters, therefore, do not cool to any extent in the few hours that it takes to traverse the canal. Since water temperatures are essentially the same exiting the discharge canal as they are leaving the plant, the shortening or removal of canal structures would reduce the duration of exposure to thermal stress encountered before effective heat dissipation begins. The elimination of the canal would probably not change the impact of the power plant on the bay area into which the effluent is discharged, because the canal structure does not greatly alter the flow or heat content of the water. Therefore, a subtropical power plant designed with little or no discharge canal would not impact an area any greater than one with such structures, but could have less entrainment effect on the copepod population.

Secondly, since salinity also plays an important role in entrainment mortality for most species, this factor should be considered in the siting of power plants. Although estuaries generally contain organisms that are adapted to survive conditions of temperature and salinity stress, considerations should be made in the siting of power plants to avoid areas in the estuarine system that are subject to fluctuations that might produce extremely low salinities that would prove lethal in combination with thermal stress (i.e., near the mouths of rivers that have a seasonal variation in flow). On the other hand, open coast siting of power plants may cause greater entrainment mortalities because open water species of copepods are less resistant to thermal stress than estuarine forms. Perhaps power plants sited on bays or sounds that are not subject to greatly fluctuating salinities would have the advantage of mainly entraining thermally resistant estuarine species while not exposing the copepods to lethal combinations of elevated temperatures and low salinities.

The fact that the majority of species are only greatly affected by certain combinations of temperature and salinity suggests that it might be environmentally profitable to monitor these factors in the power plant and adjust thermal output to avoid lethal combinations. If discharge temperatures could always be kept below 35°C, or slightly lower levels for extremely low salinities, a great deal of the entrainment mortality may be avoided. Adjusting the electrical output of the plant or pumping capacity of the cooling system could keep temperatures of the discharge waters at a level that would produce the least amount of entrainment mortality for the salinity conditions that are monitored.

Computers may prove useful tools in the monitor-control of such a system.

Laboratory Studies

Effects of Entrainment on Growth

The growth rates of Acartia tonsa from the Crystal River area were lower than those described by Heinle (1969) for the same temperatures. Zillioux and Wilson (1966) explained the differences between the growth rates that they observed and those previously reported by Heinle as being a factor of the amount of food available to the culture. Apparently, Heinle used raw sea water that contained many times the concentration of phytoplankters than would be fed to most laboratory cultures (Zillioux and Wilson, 1966). The concentration of the mixed algal diet used in the Crystal River study was intermediate between those estimated for the two studies. Since food concentration appears to be an important factor in growth, the growth rates found at Crystal River may be expected to be slightly lower in value for any given temperature when compared to those found by Heinle.

Another possible explanation of the relatively slower growth rates of Acartia tonsa populations at Crystal River compared to those of the same species in the Patuxent estuary (Heinle, 1969) is a geographic adaptation to high temperatures. Gonzalez (1974) showed that Acartia tonsa populations had different thermal tolerances depending upon the geographic location from which they were collected. If the tolerance to high temperatures of southern populations of the

species also applies to sublethal metabolic adaptations, Acartia tonsa from Florida might be expected to be less affected by temperature than northern forms. In other words, the entire metabolic activity, including growth rates, for a heat-adapted southern population may operate at a lower level than that found for a northern one at the same temperature. Such an adaptation would allow the southern form to live more efficiently at higher temperatures without being as stressed for energy (food) supply by a high metabolic rate.

Growth rates for Oithona brevicornis have not been reported. Faber (1966) reports that the instars of this species have not been previously described. Heinle (1969) did observe, however, that O. brevicornis populations in his growth experiment cultures appeared to grow at a slower rate than either the calanoid Acartia tonsa or the harpacticoid Canuella canadensis. The data from Crystal River growth studies confirm this observation, with O. brevicornis juveniles growing at nearly half the rate of A. tonsa populations.

The entrainment effects on growth are seen to be temperature related. There were no significant differences between the growth rates shown by intake or discharge populations of either Acartia tonsa or Oithona brevicornis until July. In July and every experimental period thereafter, the growth rates of the discharge populations were significantly depressed below those of the control populations. This critical period represents the dates on which the discharge canal temperatures rose above 35°C. Increases in temperature above this level appeared to cause an exponential decline in the growth rate ratio between discharge and intake populations. The exponential growth rate depression observed for rising temperatures suggests that

the process involves an Arrhenius type deactivation of substances associated with growth.

Kinne and Kinne (1962) have stated that extreme conditions of temperature, salinity or oxygen can induce developmental arrest. Such arrest can be reversible or irreversible depending on the magnitude, duration and interactions of the stressful conditions. Even if the arrest remains reversible, there appears to be an initial lag phase following normalization of the arresting conditions. The investigators attributed this lag in development to dissynchronization, lack of coordination, or insufficient speed of recoordination of developmental processes following the arrest.

Heinle (1969) described a developmental lag in an entrained population of Acartia tonsa juveniles exposed to temperatures up to 30°C. This lag in growth suggests that there was a reversible developmental arrest associated with the elevated temperatures experienced during entrainment. Of course, the Crystal River copepods, having adapted to living in a subtropical environment, might be expected to show a greater degree of thermal tolerance than that found in a northern population such as those described by Heinle (1969). This may explain why thermal effects on development did not appear to be significant until a higher temperature level was reached (35°C).

The Crystal River data seem to indicate that both reversible and irreversible developmental arrest may occur in populations of entrained copepods. From the series of histograms for each growth experiment (Appendix B), it is apparent that temperatures above 35°C cause at least a portion of the copepod population to remain in relatively early naupliar stages. This portion of the population could be assumed

to have been affected by irreversible developmental arrest. Other juveniles can be seen to regain a capacity for growth following a certain lag period. These copepods like those described by Heinle (1969), probably have been subjected to a reversible developmental arrest. The combination of reversible and irreversible developmental arrest would account for the overall depression in growth seen for Acartia tonsa and Oithona brevicornis exposed to discharge temperatures above 35°C. The dates on which temperatures were highest and growth depression greatest, a relatively larger portion of the population simply did not grow. This observation seems to suggest that reversible arrest becomes irreversible as the copepods are exposed to higher temperatures.

The relationship of salinity to entrainment-induced growth depression could not be analyzed in detail because of the relatively low numbers of observations during the period of significant effects. It was observed, however, that both Acartia tonsa and Oithona brevicornis juveniles exhibited a high growth depression during the experimental period that had the lowest salinity. A temperature-salinity interaction is assumed to have caused a greater effect on growth than expected for the temperature level (35°C). Davis and Celebrese (1964) have reported similar interactions in experiments on clam larvae, where low salinities cause reversible developmental arrest to become irreversible for the same temperature conditions. Salinity may, therefore, be as important a factor in entrainment effects on growth as it was in the initial entrainment mortality of the copepods.

Although the exact cause of developmental arrest is not known, the proceeding observations seem to indicate that certain growth substances are either reversibly or irreversibly deactivated. This deactivation proceeds in an Arrhenius type relationship with temperature and the reaction becomes increasingly irreversible with higher temperatures. Johnson et al. (1974) deal extensively with the kinetics of denaturation of enzymes and conclude that the rate at which reversible reactions become irreversible depends on the temperature and time of exposure. It appears, therefore, that growth substances are denatured at an increasingly rapid rate above a critical temperatures (35°C) and this denaturation becomes increasingly irreversible as temperatures rise above this level. These growth substances are probably enzymes that catalyze the formation of various factors needed for growth. In copepods, the growth factors may be some of the neuro-secretory hormones that induce moulting to the next developmental stage. The critical factor, on the other hand, may be so basic as to affect cell multiplication associated with growth. McWhinnie (1967) examined the literature on the biochemical mechanisms of developmental arrest in protozoans and metazoans and generalized that survival without growth in invertebrates may result from differential enzyme inactivation. He states that synthetic enzyme systems tend to be more sensitive to thermal deactivation than those involved in energy release. Therefore, an organism exposed to thermal stress may be able to feed and utilize food energy, but, at the same time, have certain enzymes involved in the synthesis of growth factors inactivated, either reversibly or irreversibly. If the enzyme system is reversibly deactivated, it will take a period of time

for the reactivation reaction to occur and for the growth factors to be synthesized at the rate they were before the thermal shock. This period of reactivation would account for the lag in growth rate observed following reversible developmental arrest.

There are several important ecological implications of the entrainment-induced growth depression. First of all, juveniles that have completely ceased to grow are effectively "dead" to further secondary production of the standing crop and might be considered as such in mortality calculations. Even the lag period associated with reversible developmental arrest may increase the turnover time of a species enough to cause lower productivity and possibly even extinction from natural mortality or predation. The decreased secondary production due to the effective growth depression could have a dramatic effect on energy flow to higher trophic levels of the ecosystem.

The overall effect of the developmental arrest is to cause the population structure of a species to shift towards the earlier stages, since the stunted juveniles remain alive but do not grow. This shift to smaller individuals compounds the trend towards smaller forms caused by differential entrainment mortality. The shift of competitive advantage to non-selective filter feeders would be accelerated by entrainment-induced growth depression increasing this trend towards smaller copepods.

The differential effects of entrainment on growth of the two species studied may have important implications. The fact that Oithona brevicornis appears to exhibit a greater amount of growth depression than Acartia tonsa may, to some degree, counteract possible

ecological effects caused by the differential entrainment mortality between the species. In other words, even though Oithona spp. appear to have a competitive advantage due to their resistance to entrainment mortality, their sensitivity to growth depression may counteract the trend or even cause the advantage to shift to the more tolerant Acartia tonsa population. Such changes in community structure and the associated ecological implications are not readily predictable because of the complexity of the subtle factors involved. A detailed calculation of the productivities of the species including all of the entrainment-induced changes would be necessary to assess the relative effects on the two species. A computer simulation of the effects would be useful in such a comparative assessment.

Temperature appears to play the major role in entrainment-induced growth depression. It must therefore be assumed that copepods passing through waters of the impacted area that have temperatures above 35°C would be subjected to nearly the same amount of growth depression as those actually entrained. If this assumption is valid, the power plant influences a great deal more organisms than actually pass through it and the subtle ecological implications may become more important to the functioning of the entire estuary.

Effects of Entrainment on Reproduction

Although the adult Acartia tonsa used in the reproductive study were subjected to laboratory conditions, the trends in reproductive rates seem to reflect quite well the reproductive activity observed in the field. Peak reproductive rates observed in the laboratory

coincided with peak densities of the species sampled in both the field and growth studies.

The results from the reproductive studies at Crystal River seem to indicate that entrainment does cause some reproductive impairment. The reproductive rates of control populations of Acartia tonsa were shown to be significantly higher than those of discharge populations. Peak periods of egg production were the times when discharge population values lagged behind those of the intake population, with significant discrepancies coming during the hottest months (discharge temperatures above 35°C).

The trend for the discharge population to lag behind during peak periods of egg production may mean that the trauma of entrainment has caused the copepods to cease reproduction for some period of time following entrainment. The total number of eggs that are produced once egg production resumes may never match that of the control population which continued to produce at high rates. If, however, both populations are producing eggs at low rates, the discharge population may be able to catch up to the control population in numbers of eggs during the experimental period.

The lower reproductive rates for the discharge population could also be due to a metabolic response to thermal stress. The increase in metabolic activity associated with the initial thermal shock reaction (Jensen et al., 1969) may utilize oxygen and nutrients that would have otherwise gone into the process of egg production. This would allow control cultures of Acartia tonsa to get a head start on the egg production, while discharge populations adapted metabolically and renewed nutrient reserves. In fact, Fox and Phear (1953) have

suggested that certain cladocerans cut back on egg production at high temperatures as an adaptive mechanism against anoxia to prevent excess drain of oxygen to the developing eggs. The thermal stress encountered in entrainment may have caused a similar shunting of energies away from reproductive activity in the A. tonsa adults.

A third possible explanation of the depression in reproductive rate is that the heat had some effect in altering hormones, enzymes, or biochemical substances necessary in reproductive activities. The exponential rate at which the reproductive depression occurs with increases in temperature above 35°C suggests that this process is also the result of an Arrhenius type denaturation. The inactivation of any substances that are involved in gametogenesis or other reproductive activity would result in the depression in reproductive rates. There is no reason to believe that developmental arrest, either reversible or irreversible, could not occur in some or all of the developing gametes within the adults. Inactivation of a synthetic enzyme system could slow or stop the process of gametogenesis, because certain critical developmental substances could not be formed. On the other hand, inactivation of some of the hormones controlling reproductive activities could also delay reproduction until new ones can be produced. Jensen et al. (1969) state that these neuro-endocrine transmitter substances may be the most thermally vulnerable protein complexes of an adult invertebrate. It must be noted that very few discharge cultures showed a complete absence of egg production. This would indicate that the thermal inactivation either involved substances that can be renewed, is for the most part reversible, or only

involves a certain percentage of the adult copepods' store of the critical substances within the range of temperatures studied.

Whichever of these or other possible explanations are true, the overall effects remain the same: the discharge populations of Acartia tonsa have a depressed reproductive capacity over the 5 to 7 days of the experiment and probably for the remainder of their rather short life span. This reproductive depression could have a significant effect on the standing crop and secondary production of the area. In terms of the production of a new generation of Acartia tonsa, the decreased reproductive capacity has the same effect as if a larger portion of the adult population had been initially killed by entrainment. Therefore, this subtle effect of entrainment could have as great an impact on the production of an area as would the obvious effect of increased entrainment mortality. It might be further speculated that changes in community structure and energy flow patterns could occur if there is differential reproductive depression between the species of copepods of the area.

As with the other entrainment effects, depression of reproductive rate appears to be a function of discharge temperatures. Water temperatures above 35°C may, therefore, cause depression in the reproductive capacity of copepods entering the impacted area without being entrained. The scope of concern would include many more organisms than are actually passing through the power plant and the secondary production of an extended area of the estuary may be affected during the summer months.

Effects of Entrainment on Long-Term Survival

Juvenile Acartia tonsa and Oithona brevicornis tested for long-term survival during the growth study experiments showed no significant differences between intake and discharge populations. Even during extreme summer conditions where growth depression was observed, the survival of both populations were shown to be similar. This indicates that long-term lethal effects are not apparent in juveniles that have survived the initial 6 to 8 hours following entrainment, at which time growth experiments were started.

Similarly, adult Acartia tonsa from intake and discharge areas showed no significant differences in long-term survival during the reproductive study. This appears to indicate that, if the adult copepods survive the first 24 hours following entrainment, no further lethal effects are seen.

Culturing temperature appears to play a significant role in determining the mortality rate for adult copepods, while it does not for the juveniles. This would seem to indicate that the mortality rate of the adults is largely controlled by aging and natural senescence. The rate of these processes has been shown to be a function of temperature for many invertebrates (Strehler, 1962). In the cultures of juveniles copepods, on the other hand, increases in temperature accelerate the growth rates, but death due to old age is not seen within the time frame of the experiments. The temperature-controlled aging process would therefore not greatly influence the mortality rate of juveniles.

Strehler (1962) studied the relationship between thermal shock and long-term survival in Drosophila and observed that individuals that survived the short-term effect of thermal shock did not subsequently show any increased probability of death over that of controls maintained at constant temperatures. He concluded that factors producing death in Drosophila as a result of normal senescence were not related to processes involved in thermal death. The data from the Crystal River study indicate that similar observations can be made for the copepod Acartia tonsa. Individuals of this species that survive the thermal stress of entrainment show no higher probability of mortality than controls that are maintained at intake canal temperatures. The processes of thermal death and natural mortality for this species appear unrelated. This assumption can be visually confirmed by comparing the nearly linear relationship of natural mortality to temperature shown in Figure 60 to the steep exponential rise in entrainment mortality shown for temperatures above 35⁰C in Figure 10a. The dramatic differences in the slopes of these curves indicate that two unrelated processes, with different activation energies and different relationships to temperature, are taking place.

SECTION V

SUMMARY AND CONCLUSION

1. A study was made of the immediate lethal effects, as well as the subtler biological aspects, of power plant entrainment and the associated thermal stress on the copepods of the Crystal River estuary. Both types of entrainment effects are seen to have important biological and ecological implications.
2. The copepod species of the Crystal River area appear to be representative of those found in previous studies on various estuaries in Florida and the southeastern United States. In terms of mean numbers of individuals per cubic meter, the important species listed in order of abundance were Oithona spp., Acartia tonsa, Paracalanus crassirostris, Euterpina acutifrons, Pseudodiaptomus coronatus, Tortanus setacaudatus and Labidocera spp.
3. The temperature of the heated effluent and the duration of exposure to the elevated thermal conditions of the discharge canal were the most significant factors in copepod mortality. Mortalities were generally low for discharge temperatures below 30°C, rose to moderate levels between 31 and 35°C, and increased in an exponential manner between 35 and 37°C. This trend indicates that the time of most concern for lethal entrainment effects would be for days that the discharge temperatures climb above 35°C.

4. Although all of the copepod species appear to be living near their upper thermal limits during the hottest months, some appear to be living much closer to this critical level and are more sensitive to relatively lower discharge temperatures. In general, the true estuarine species appear to be better adapted to thermal stress and have higher thermal limits than the coastal species.
5. There appears to be a temperature-salinity interaction where conditions of low salinities and high temperatures or high salinities and low temperatures cause increased entrainment mortality. On the other hand, high salinities seem to "buffer" the effects of moderately high temperatures for most species of copepods.
6. Mechanical damage caused by passage through the power plant seems to account for a rather small percent mortality, but may be the major lethal factor in entrainment during the colder months. The size of the copepod seems to be an important factor in determining the amount of damage caused by the mechanical aspects of entrainment.
7. In general, juvenile copepods seem to survive entrainment much better than adults, and males seem more sensitive to entrainment effects than females.
8. The effects of entrainment on growth of juvenile copepods appear to be related to temperature. Growth rates for entrained populations of Oithona brevicornis and Acartia tonsa were depressed below those of control populations during the hottest months (July-September). Over this period, entrained Acartia tonsa grew about 54% slower than intake individuals. Oithona brevicornis

juveniles taken from discharge waters showed a 75% depression in growth rates below those of intake populations during these months.

9. The entrainment-induced growth depression appears to be the result of a developmental arrest of a portion of the copepod population. The arrest may be reversible, but appears to become increasingly irreversible at high temperatures. The stunting of growth of a portion of the entrained population may have the effect of decreasing the overall average size of copepods in the discharge area, which could have important ecological implications.
10. Temperature appears to be an important factor in the effects of entrainment on reproduction of copepods. The reproductive capacity of discharge populations of Acartia tonsa was depressed below that of intake populations during the summer months. The reproductive rates of the discharge population during this period was, on the average, 41% below that of the intake population. Significant reproductive depression was only observed for this period of high discharge temperatures (above 35°C).
11. The long-term survival of copepods that survived initial entrainment effects was not significantly different from that of control populations. The same trend was shown for copepod nauplii and copepodites with populations that have survived the first hours following entrainment showing a daily mortality rate not significantly different from control populations.
12. The mortality rate attributable to aging processes in adult copepods is significantly influenced by temperature, with higher culture temperatures causing a higher mortality rate and a shorter

life span. The processes involved in mortality due to senescence do not, however, appear to be closely related to those causing thermal death.

13. All of the significant entrainment effects showed an Arrhenius type relationship to temperature. This trend suggests that thermal denaturation of critical substances is the underlying cause of most of the lethal and sublethal entrainment effects on copepods.
14. The relationship between temperature and the important entrainment effects suggests that a great deal more of the copepod population may be affected by the thermal stress than those actually passing through the cooling system of the power plant. All copepods that are tidally recruited into the thermally impacted area during the summer months may suffer most of the same lethal and sublethal effects as an entrained population. The scope of concern could include a much larger portion of the copepod population of an estuary and, considering the short residence time of water masses in most estuarine basins, possibly even of the surrounding coastal area. The entrained organisms could therefore represent only a small portion of the biota that could be affected by the power plant during the summer months.
15. Thermally induced copepod mortality, as well as depression of growth and reproduction, could have important implications for the secondary productivity and standing crop of the estuary. In the summer, the reduction of productivity due to the subtle effects would tend to work in concert with the direct loss due to increased entrainment mortality and could possibly mean that

the biomass available to the higher trophic levels would fall below critical levels. The implications of such a deficit to the food web of the estuary could obviously be far reaching.

16. Subtle changes in community structure and energy flow patterns could result from the differential response to entrainment seen for the various species of copepods. The relative shifting of energy to passive filter feeders, to the benthic community, or to decomposers, are just a few of the possible changes in energy flow that could occur in the estuary as an indirect result of the artificial selection pressure imposed by the power generating plant.

APPENDICES

APPENDIX A

Growth Curves

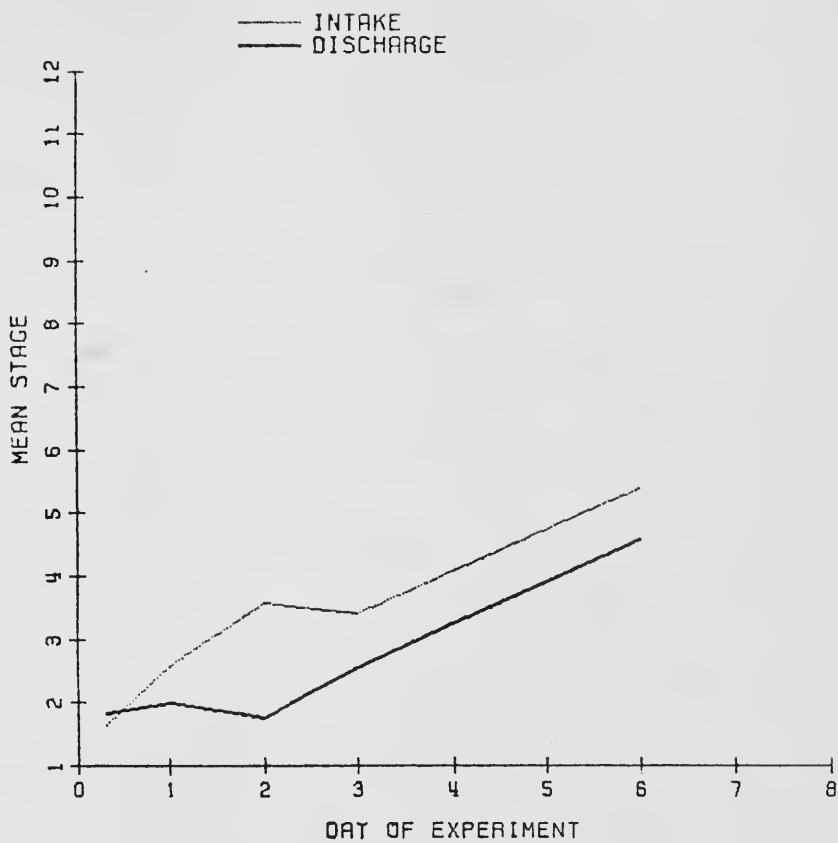


Fig. A-1. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on November 13, 1973. Mean development stage vs. day of growth experiment.

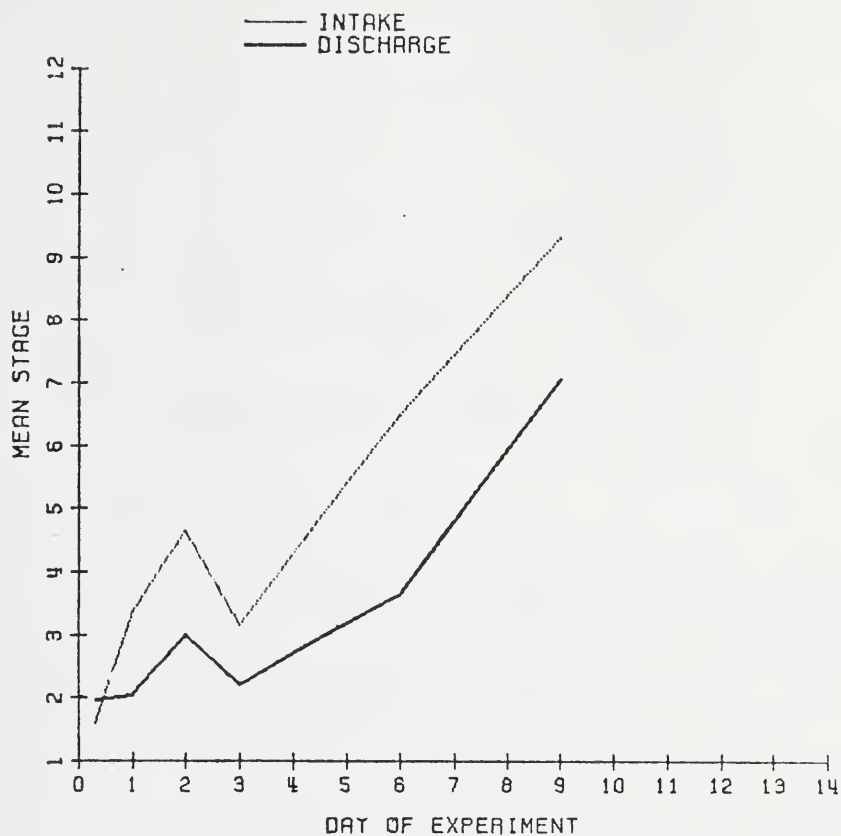


Fig. A-2. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on November 27, 1973. Mean development stage vs. day of growth experiment.

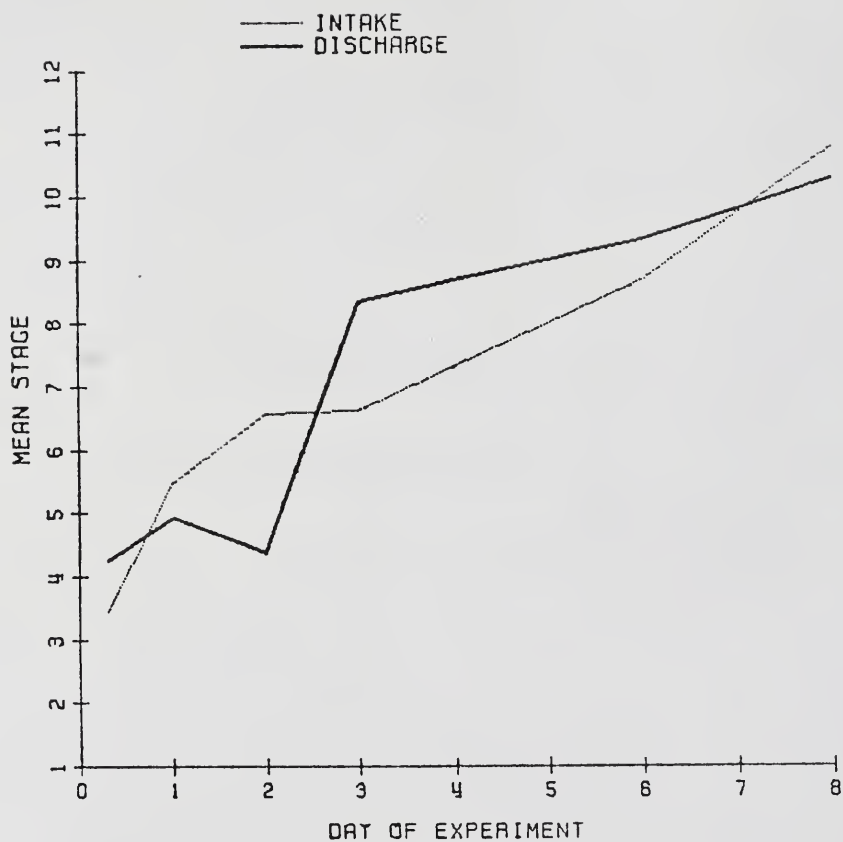


Fig. A-3. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on December 26, 1973. Mean development stage vs. day of growth experiment.

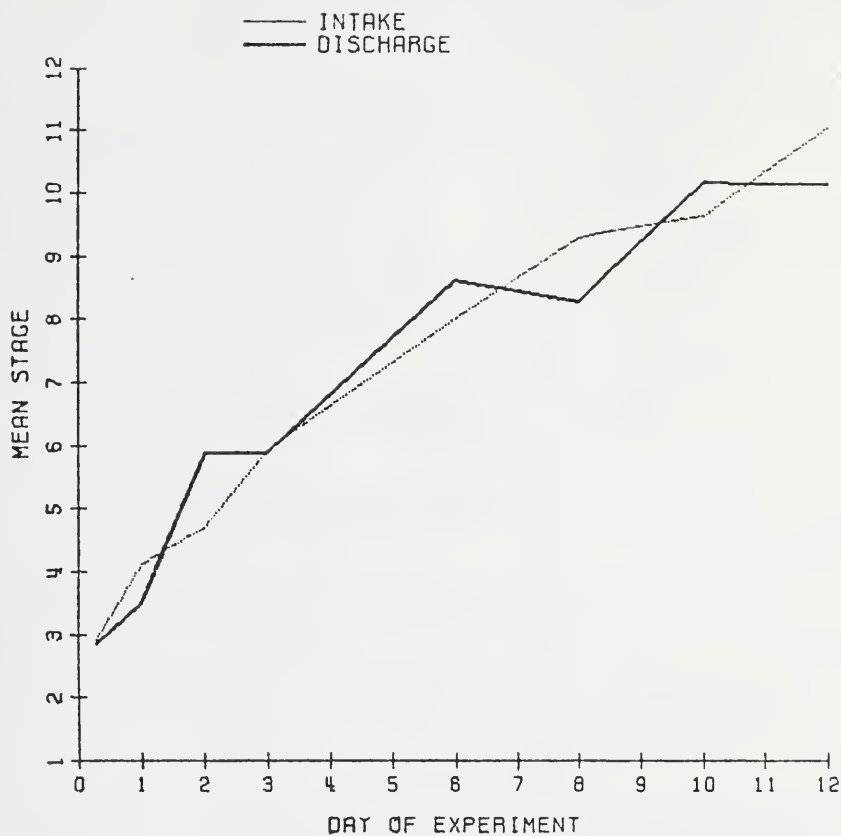


Fig. A-4. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on January 22, 1974. Mean development stage vs. day of growth experiment.

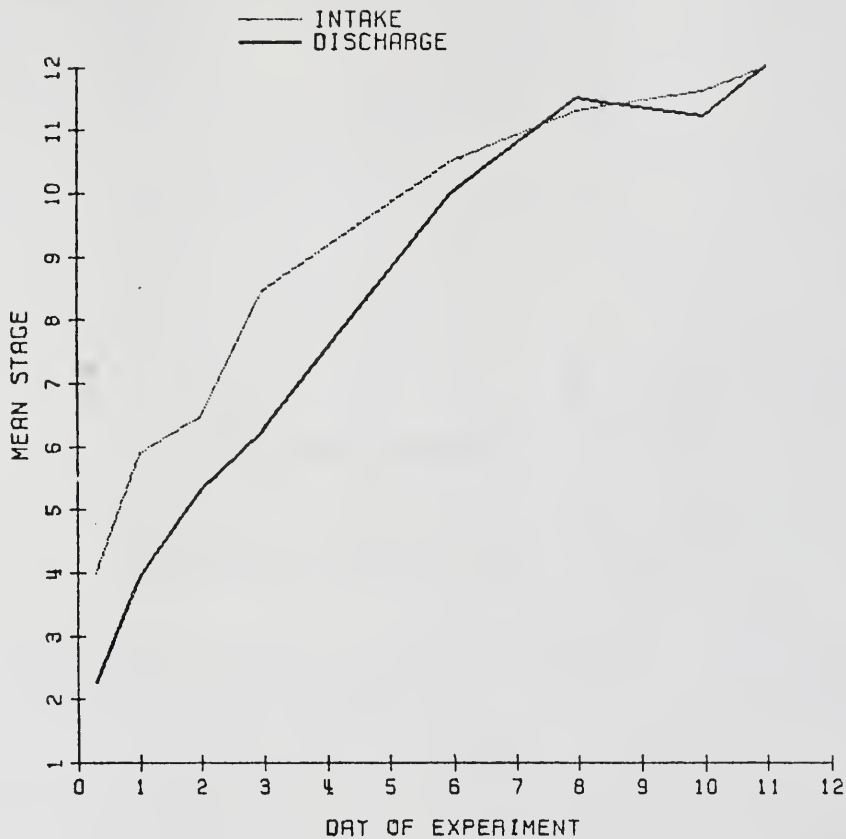


Fig. A-5. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on February 5, 1974. Mean development stage vs. day of growth experiment.

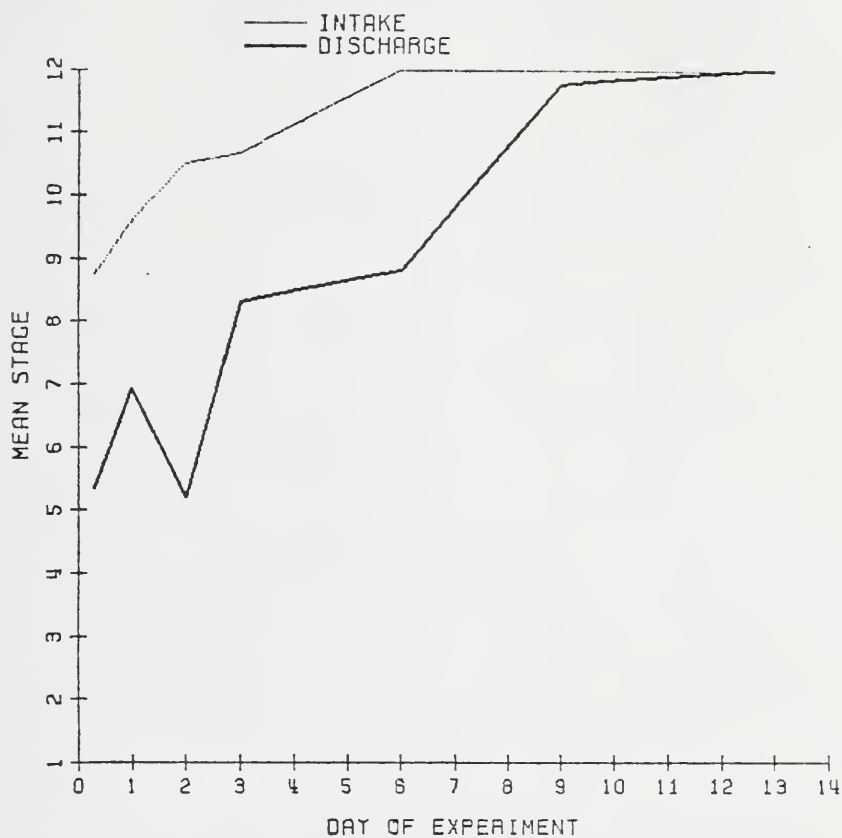


Fig. A-6. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on March 19, 1974. Mean development stage vs. day of growth experiment.

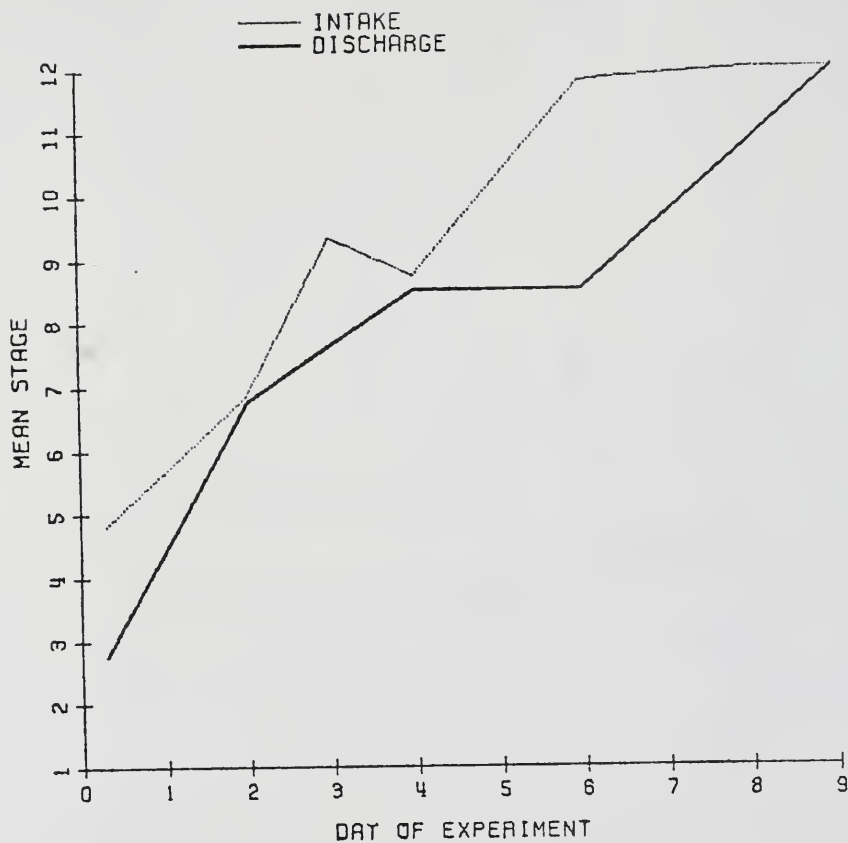


Fig. A-7. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on May 28, 1974. Mean development stage vs. day of growth experiment.

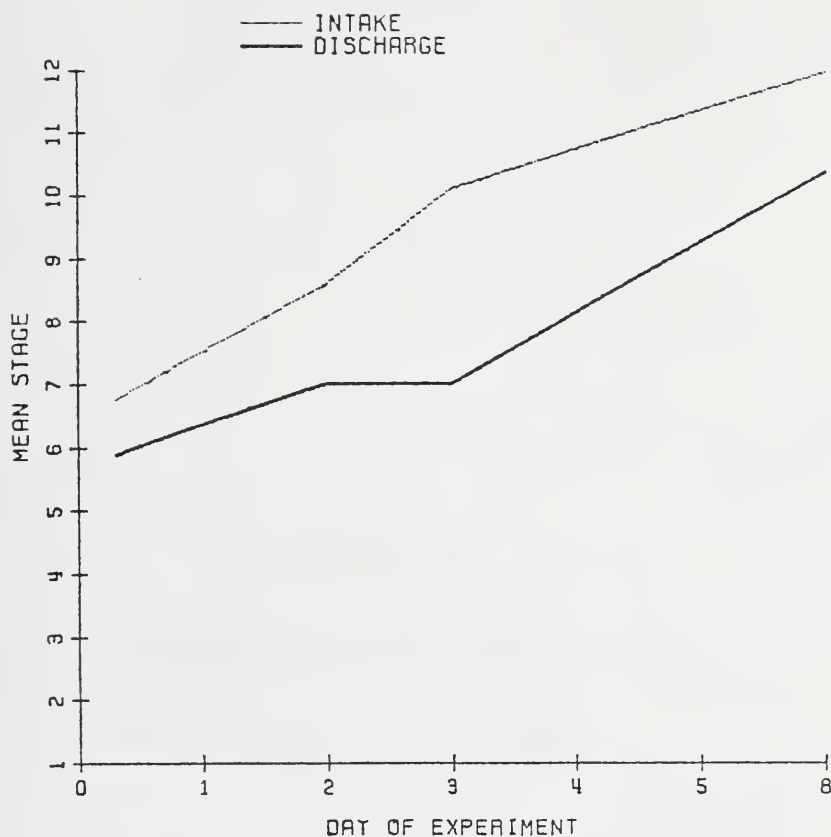


Fig. A-8. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on June 11, 1974. Mean development stage vs. day of growth experiment.

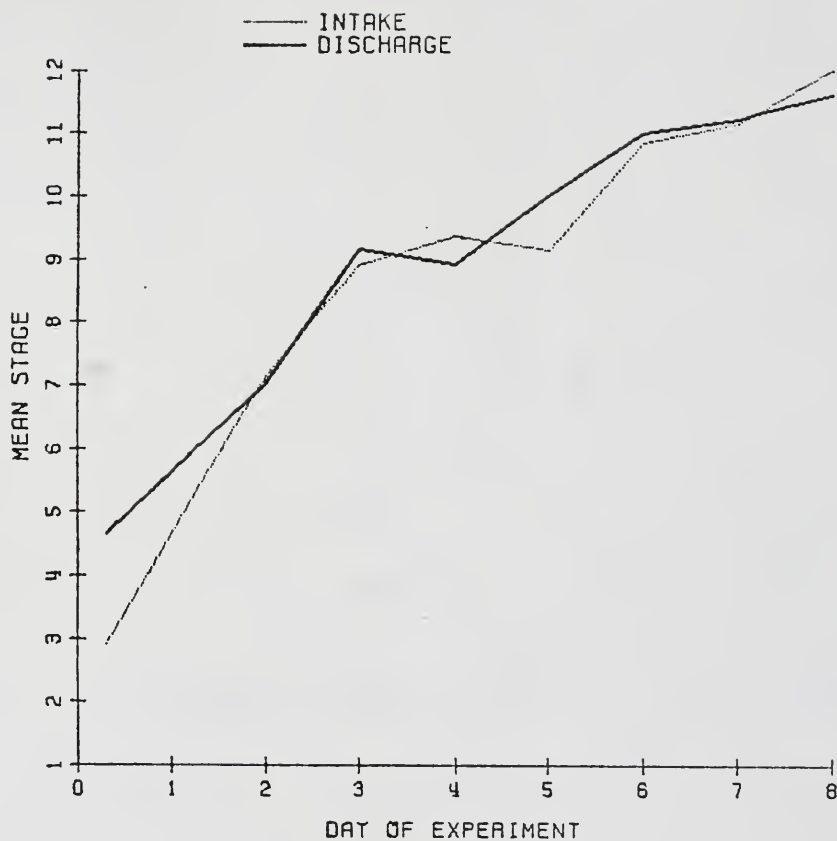


Fig. A-9. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on June 25, 1974. Mean development stage vs. day of growth experiment.

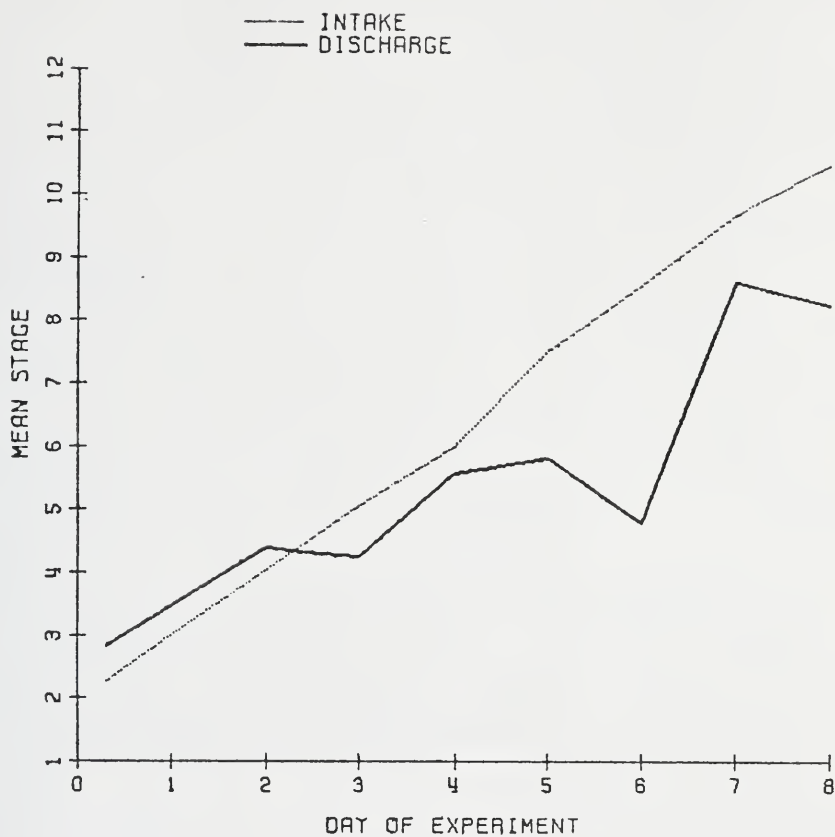


Fig. A-10. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on July 9, 1974. Mean development stage vs. day of growth experiment.

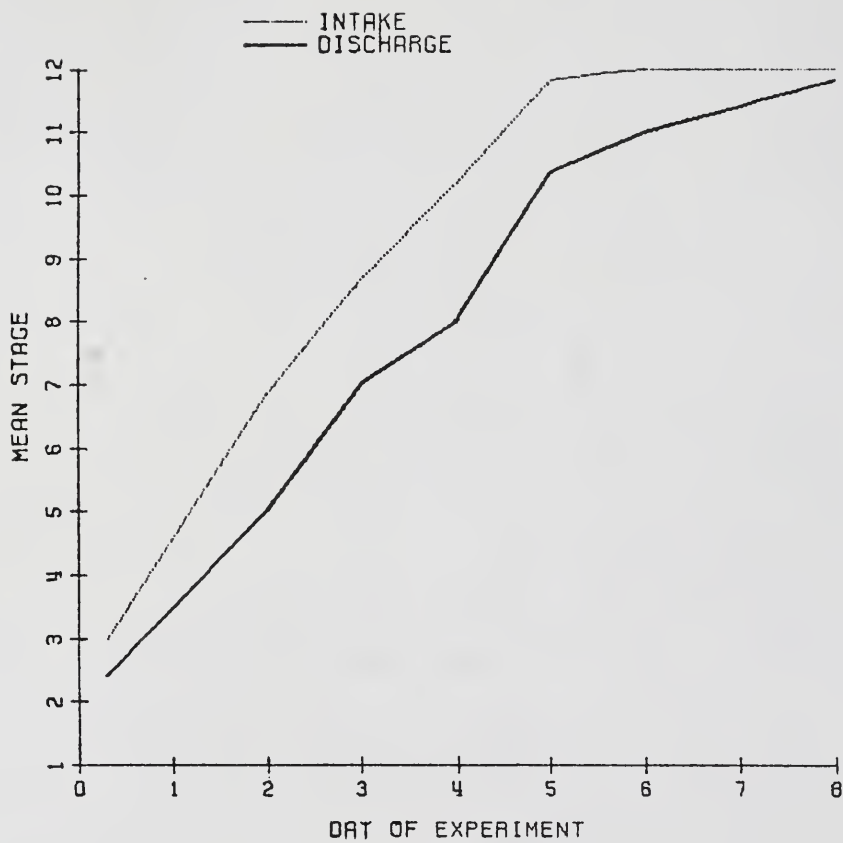


Fig. A-11. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on July 23, 1974. Mean development stage vs. day of growth experiment.

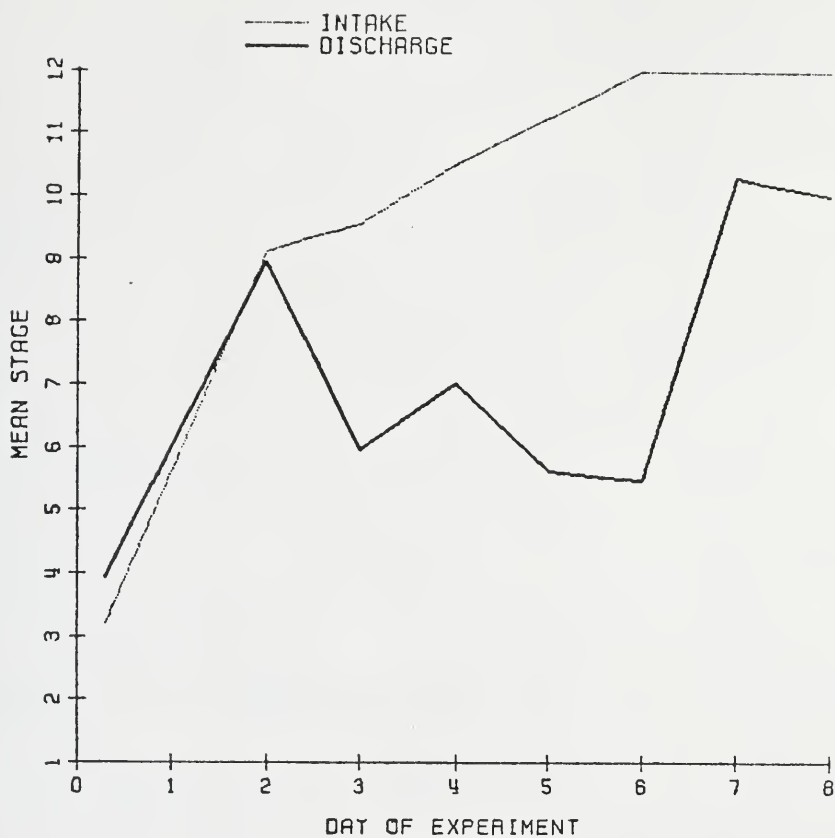


Fig. A-12. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on August 6, 1974. Mean development stage vs. day of growth experiment.

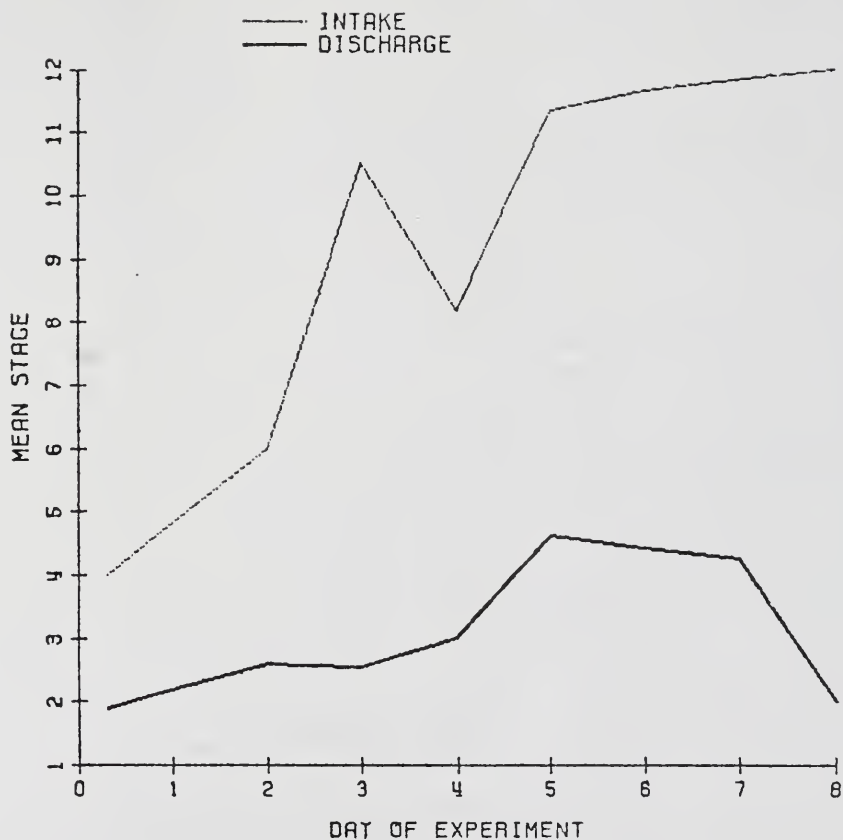


Fig. A-13. Growth curve for intake and discharge population of *Acartia tonsa* juveniles collected on August 20, 1974. Mean development stage vs. day of growth experiment.

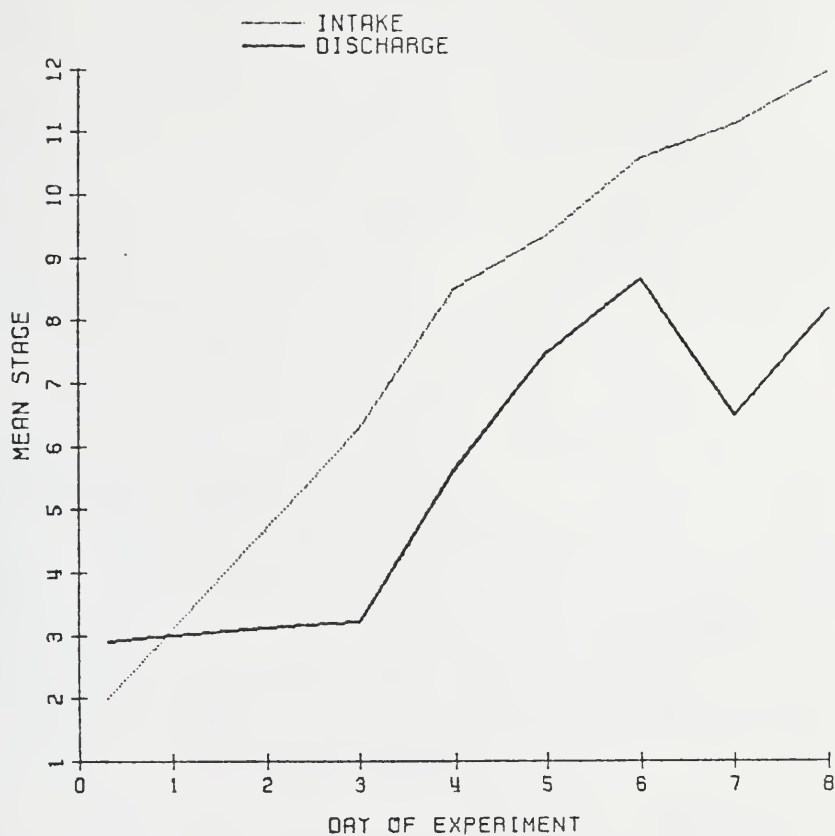


Fig. A-14. Growth curve for intake and discharge populations of *Acartia tonsa* juveniles collected on September 3, 1974. Mean development stage vs. day of growth experiment.

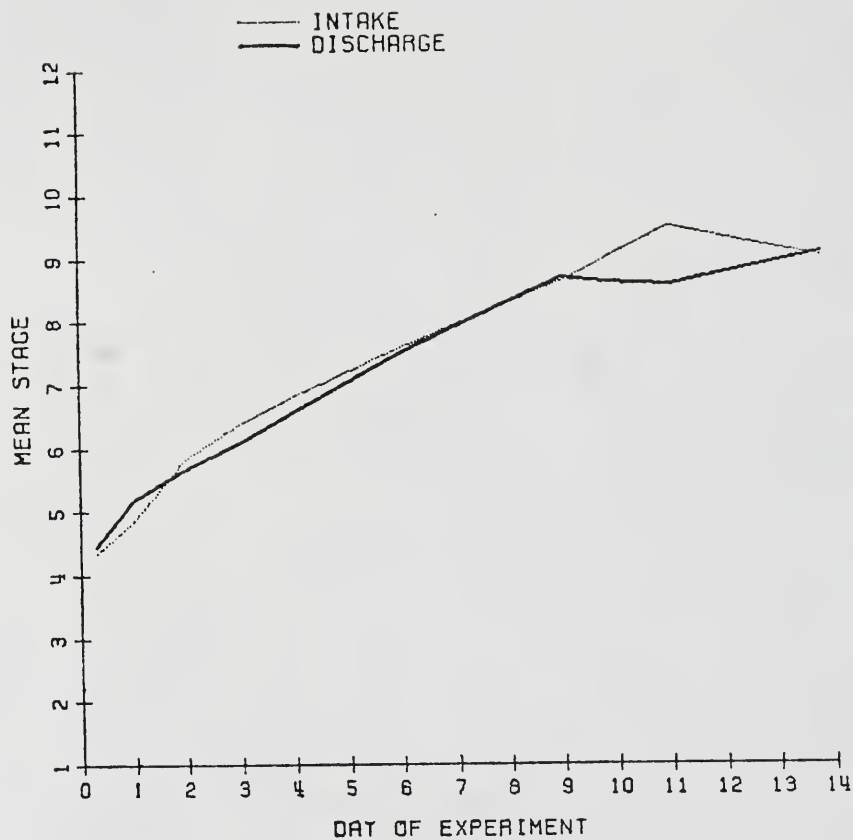


Fig. A-15. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on November 27, 1973. Mean development stage vs. day of growth experiment.

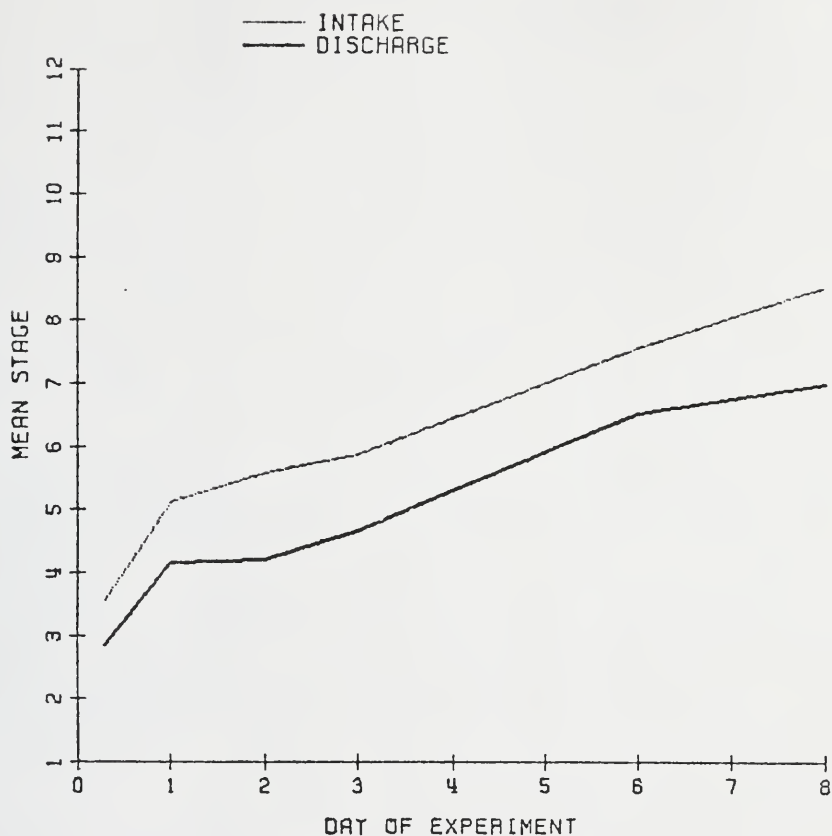


Fig. A-16. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on December 26, 1973. Mean development stage vs. day of growth experiment.

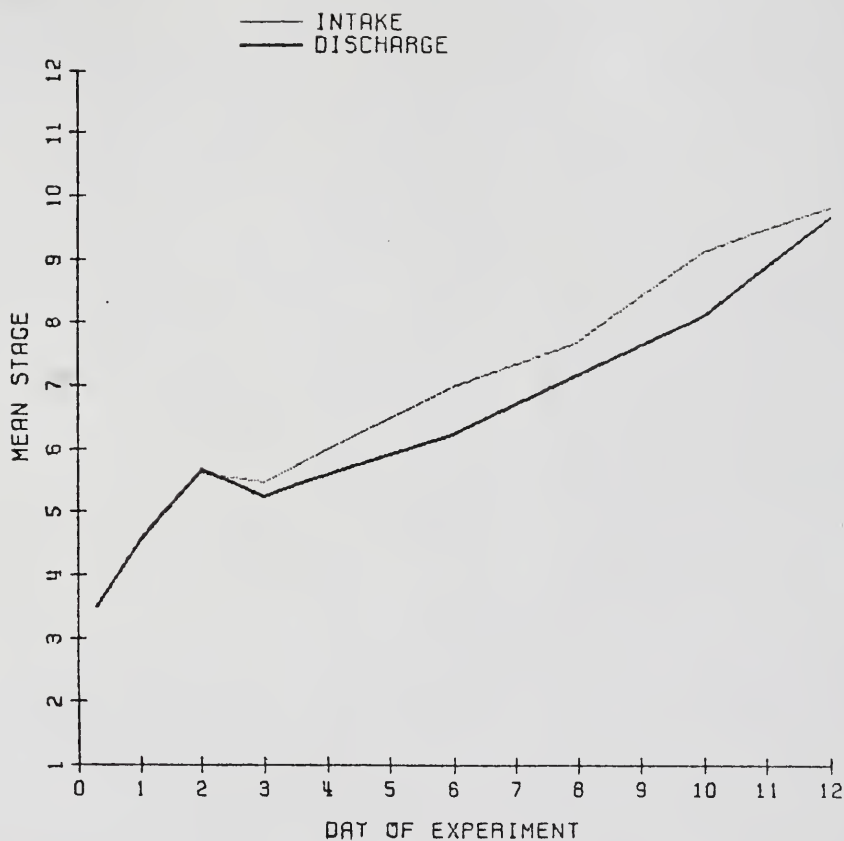


Fig. A-17. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on January 22, 1974. Mean development stage vs. day of growth experiment.

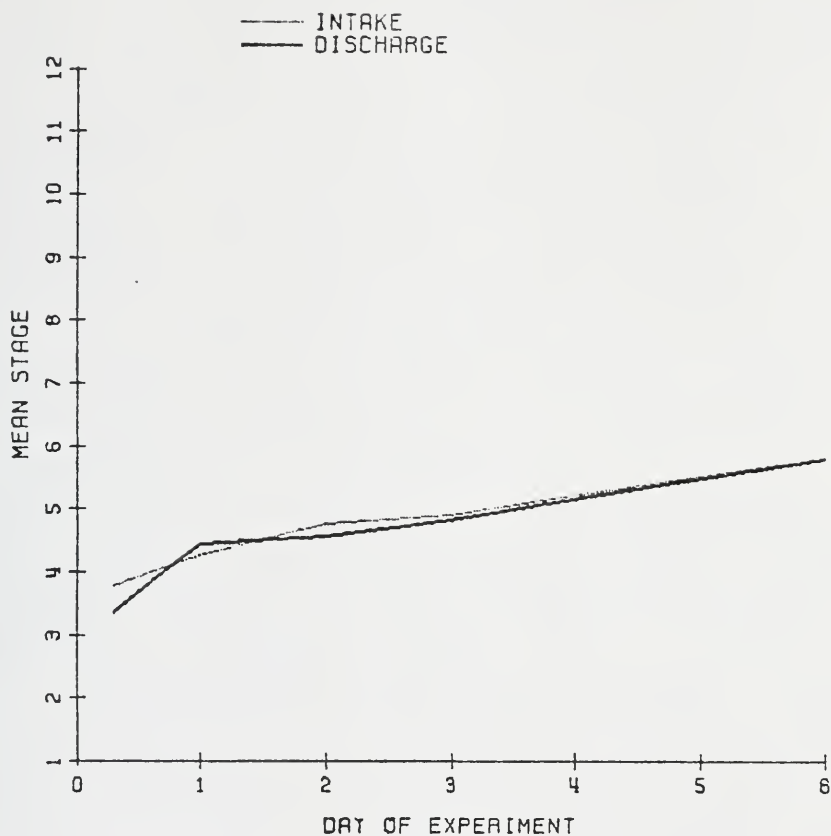


Fig. A-18. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on February 5, 1974. Mean development stage vs. day of growth experiment.

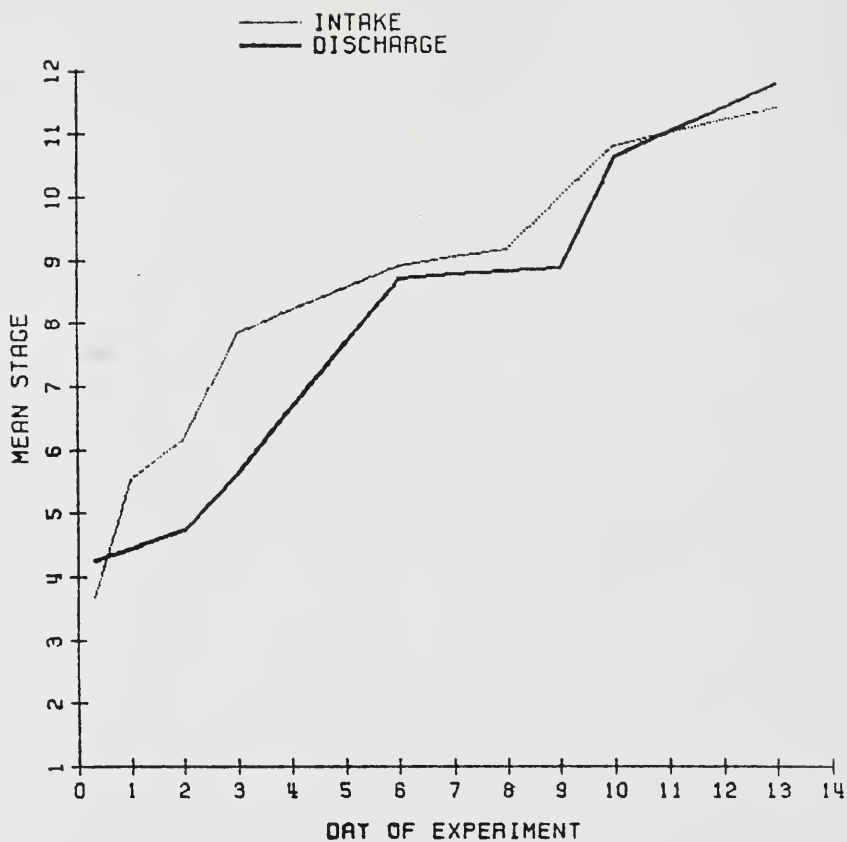


Fig. A-19. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on March 19, 1974. Mean development stage vs. day of growth experiment.

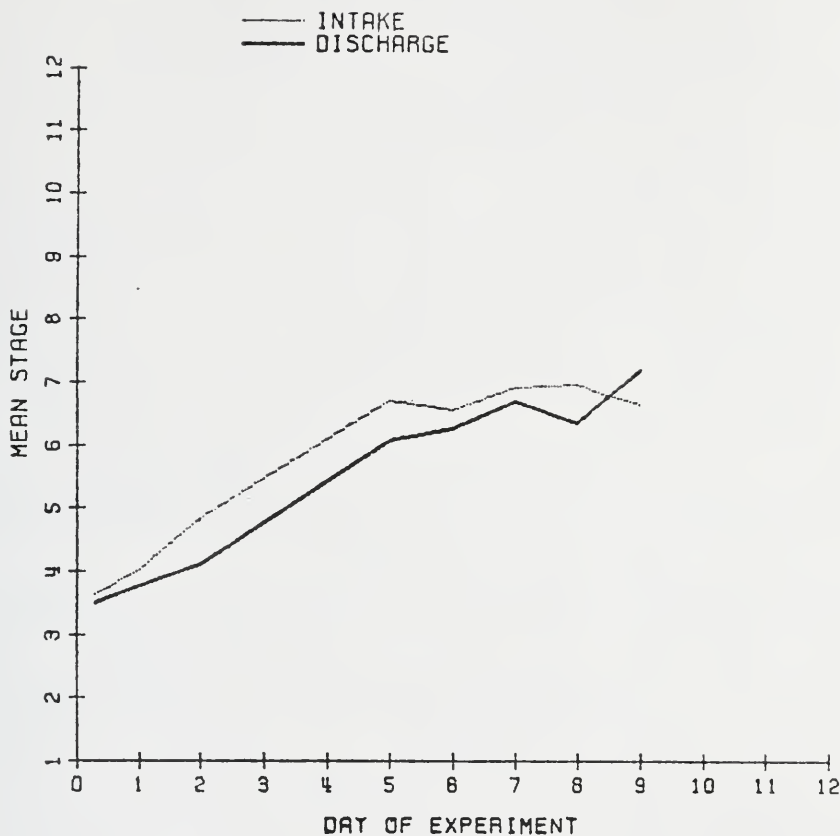


Fig. A-20. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on April 24, 1974. Mean development stage vs. day of growth experiment.

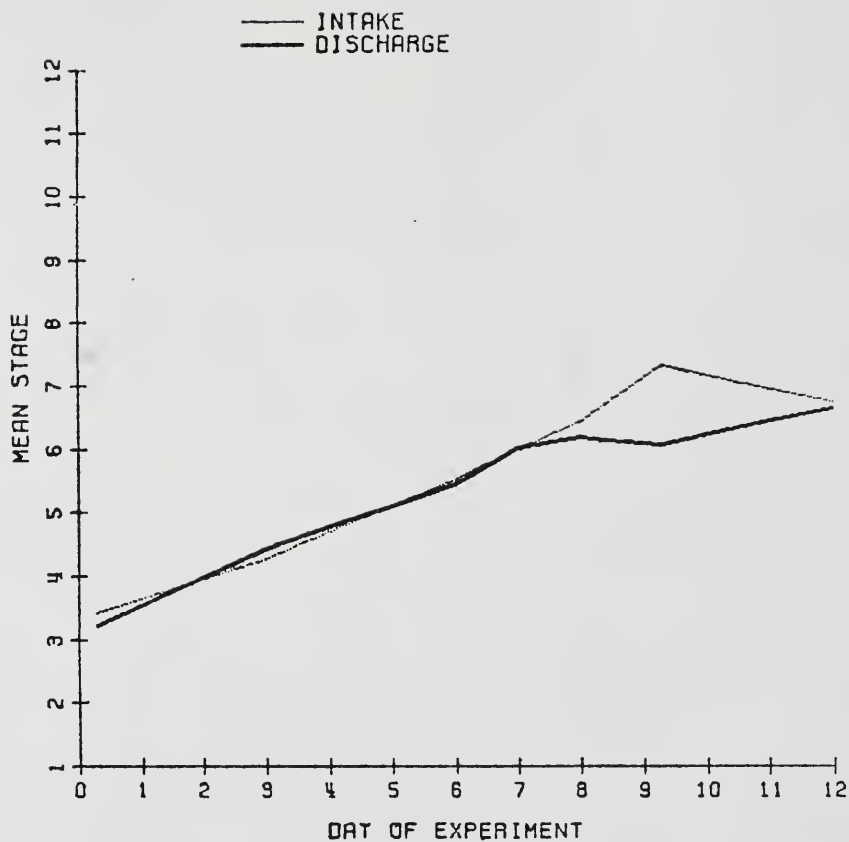


Fig. A-21. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on May 14, 1974. Mean development stage vs. day of growth experiment.

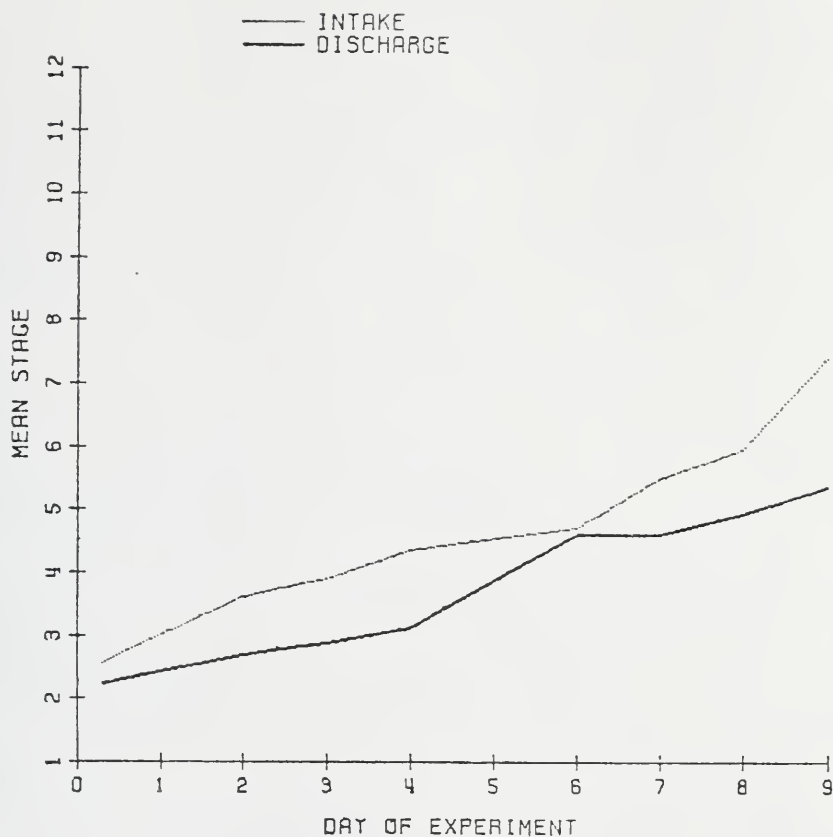


Fig. A-22. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on May 28, 1974. Mean development stage vs. day of growth experiment.

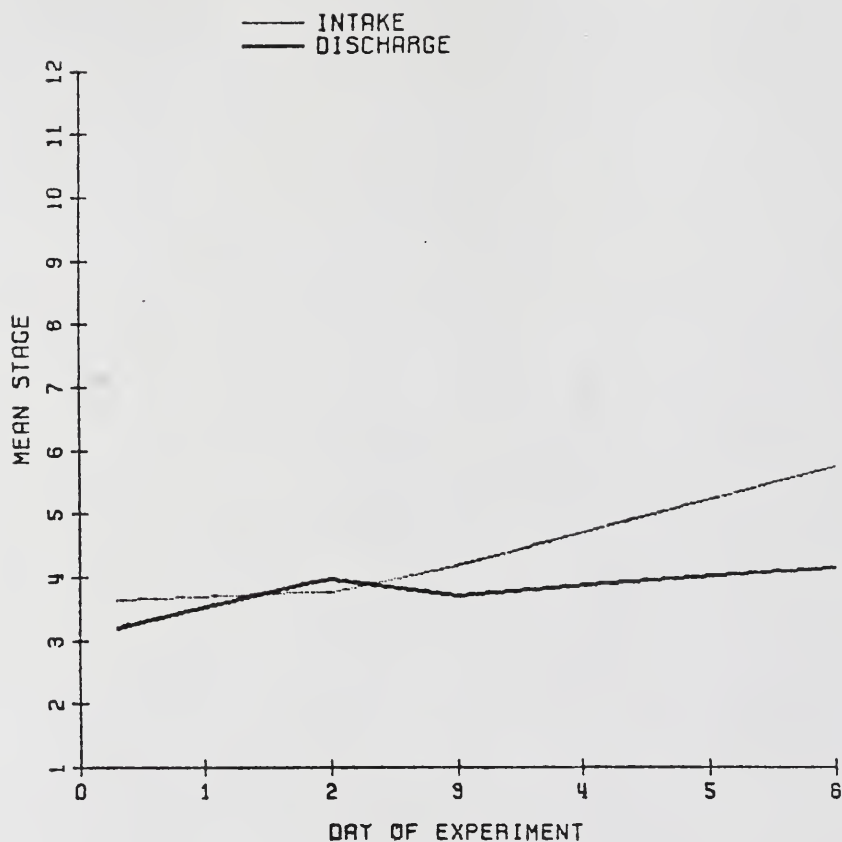


Fig. A-23. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on June 11, 1974. Mean development stage vs. day of growth experiment.

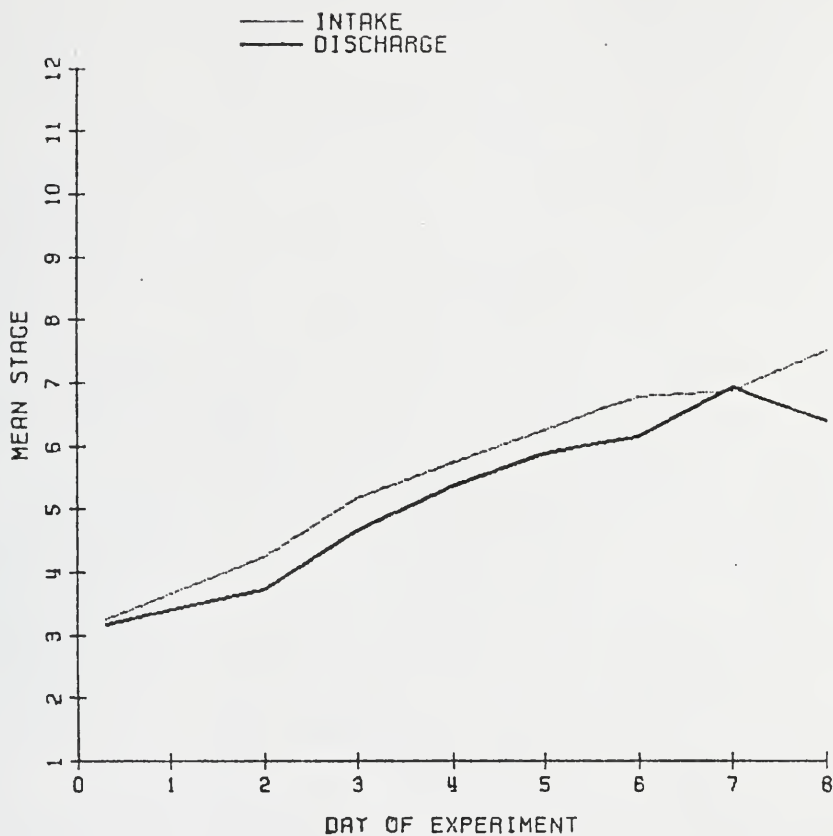


Fig. A-24. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on June 25, 1974. Mean development stage vs. day of growth experiment.

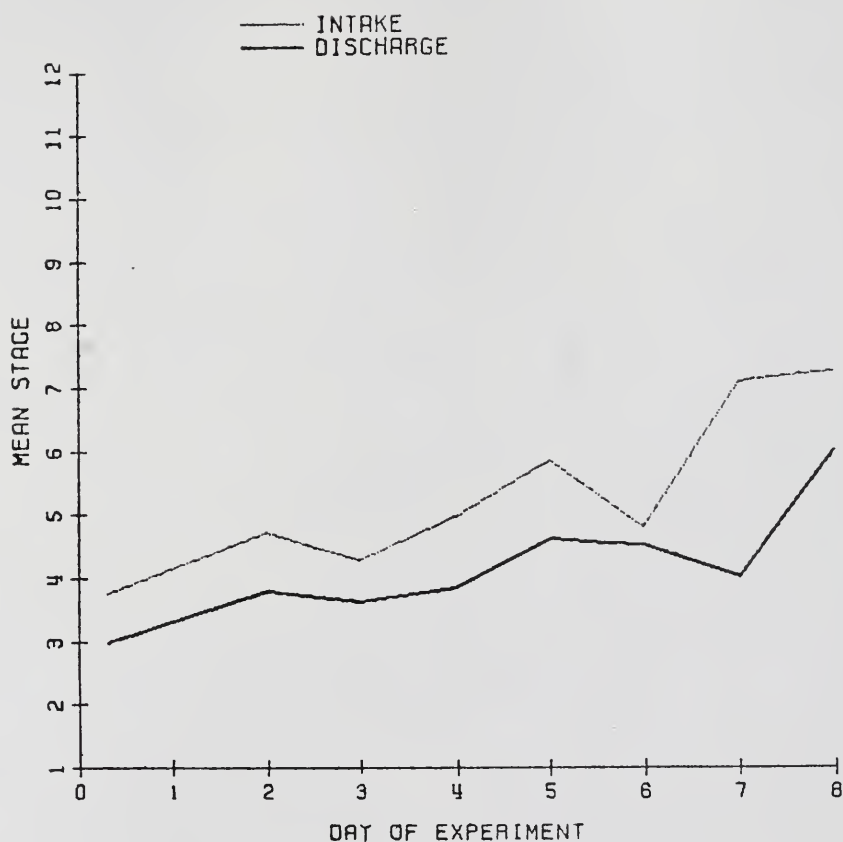


Fig. A-25. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on July 9, 1974. Mean development stage vs. day of growth experiment.

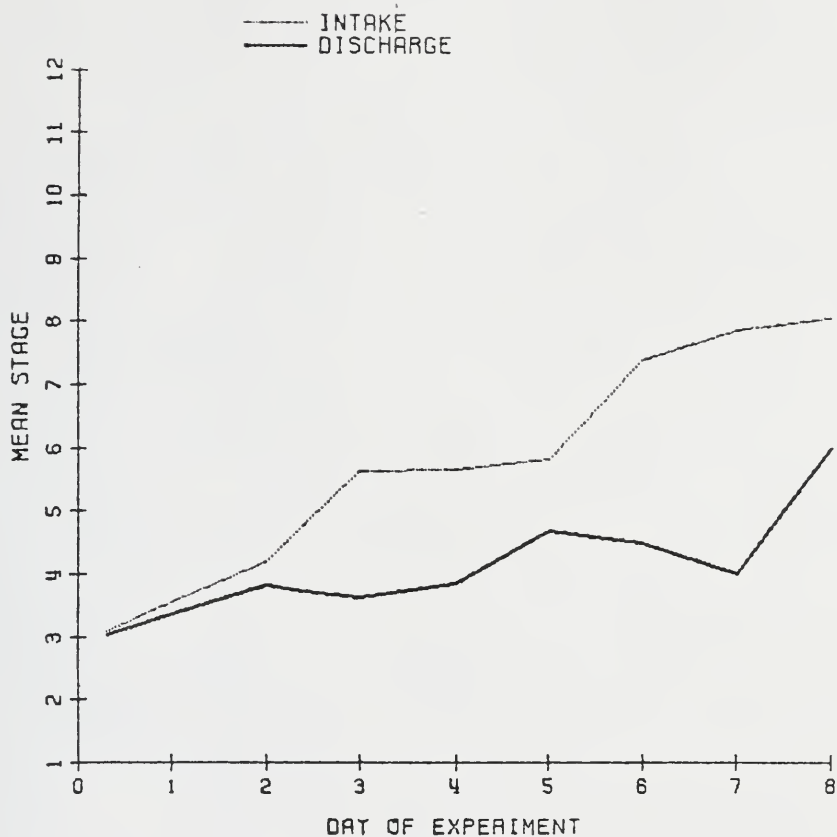


Fig. A-26. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on July 23, 1974. Mean development stage vs. day of growth experiment.

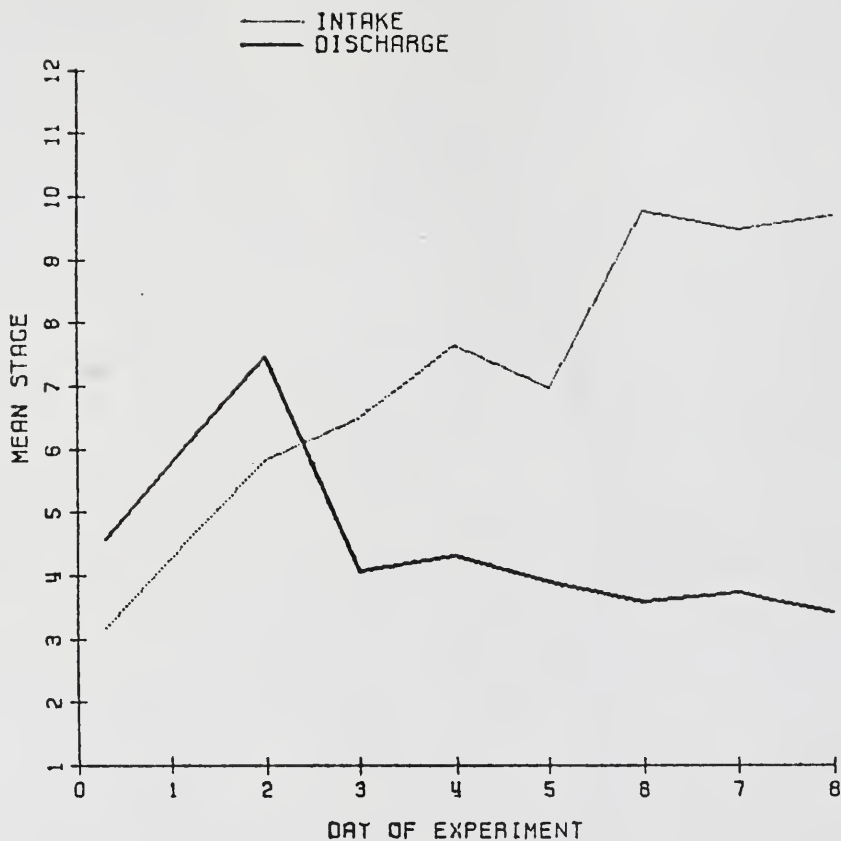


Fig. A-27. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on August 6, 1974. Mean development stage vs. day of growth experiment.

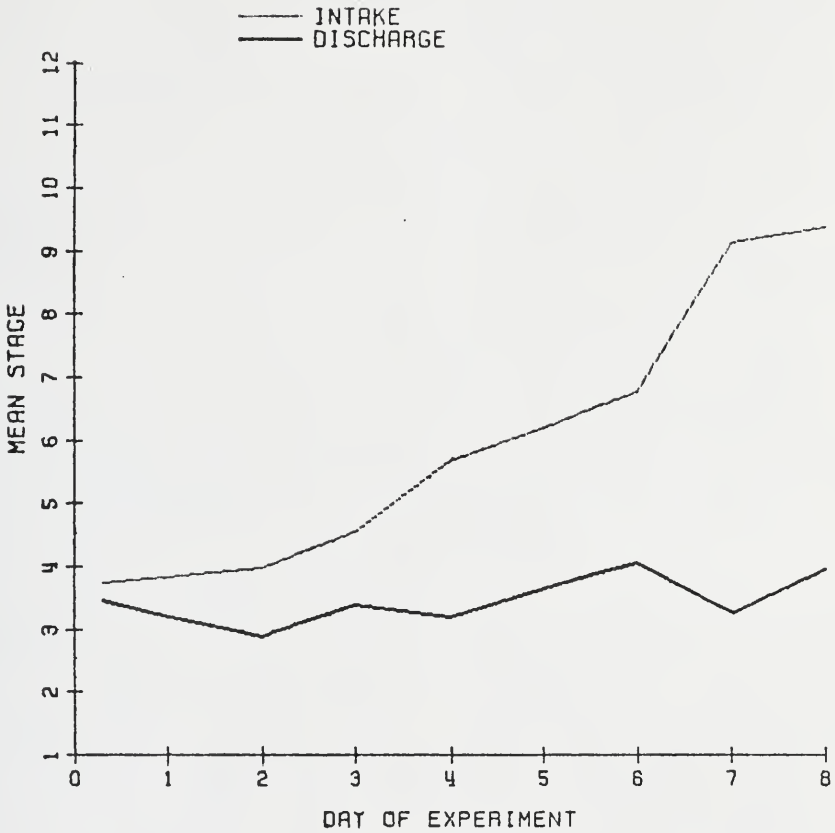


Fig. A-28. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on August 26, 1974. Mean development stage vs. day of growth experiment.

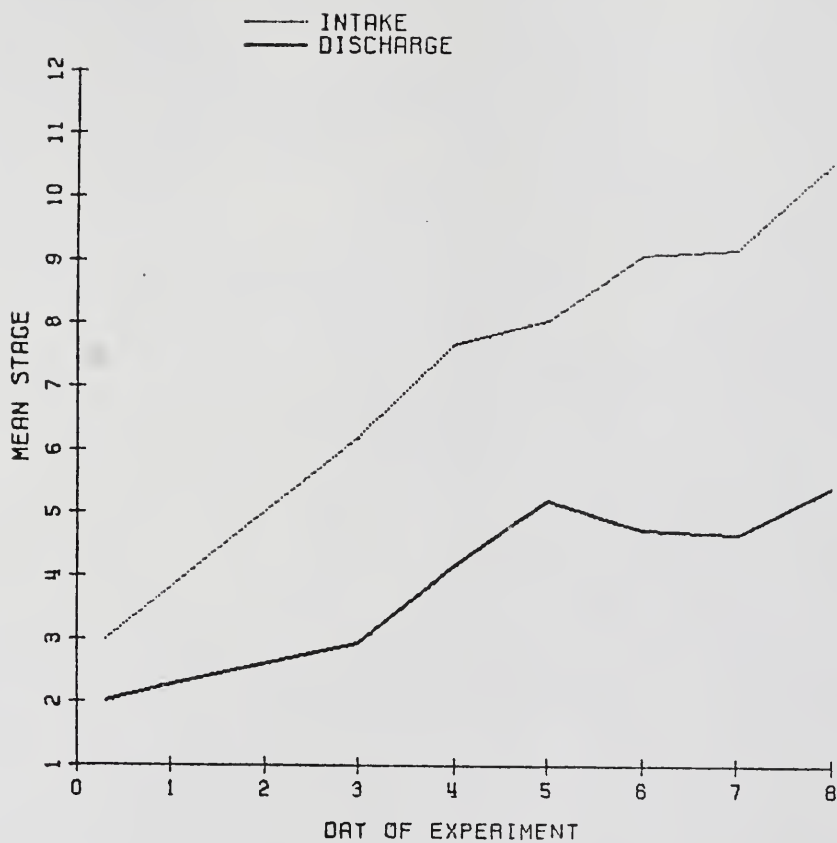


Fig. A-29. Growth curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on September 3, 1974. Mean development stage vs. day of growth experiment.

APPENDIX B

Population Age Structure Histograms

N1 to N6 are the six naupliar stages, while C1 to C5 are the five copepodite stages, C6 is the adult.

Fig. B-1. Histogram series for Acartia tonsa in growth experiment started on November 13, 1973, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 3, (e) Day 6.

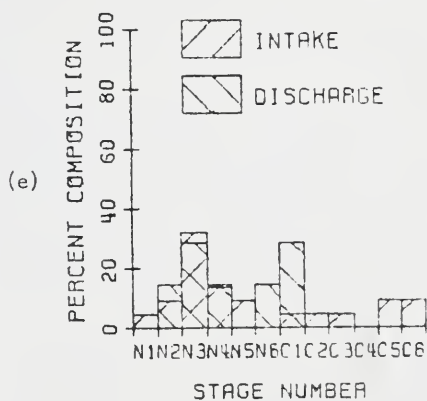
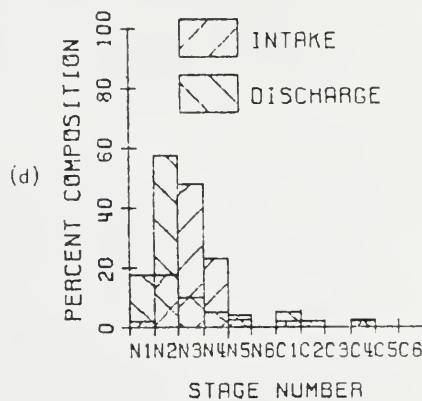
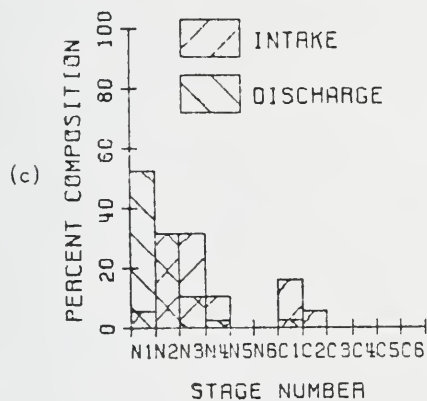
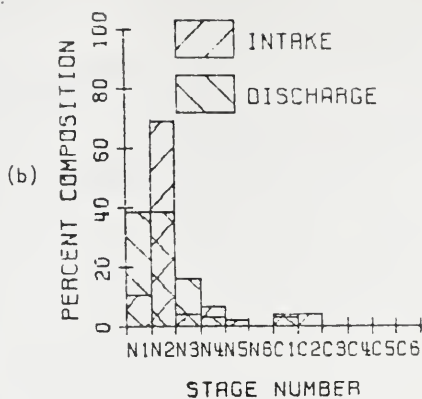
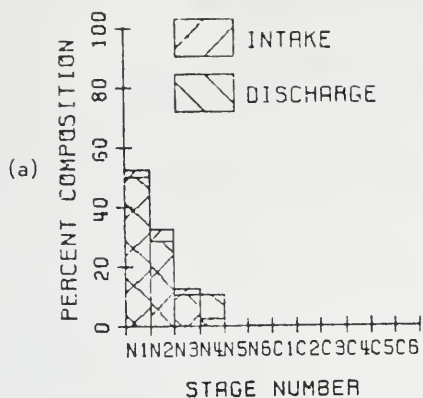


Fig. B-2. Histogram series for Acartia tonsa in growth experiment started on November 27, 1973, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 3, (e) Day 6, (f) Day 9.

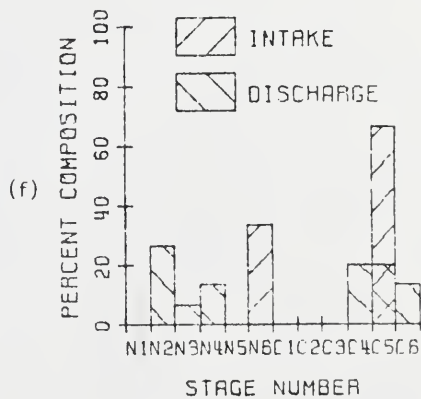
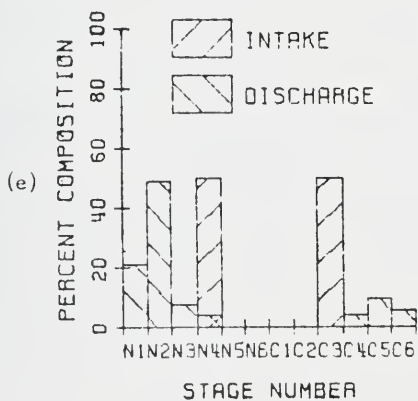
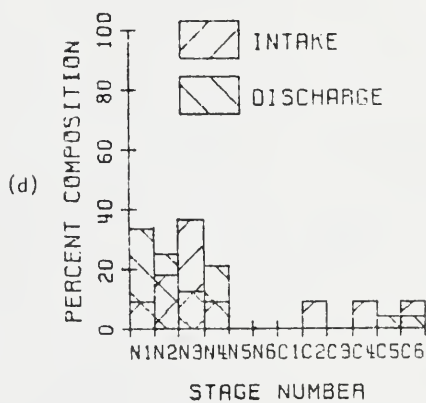
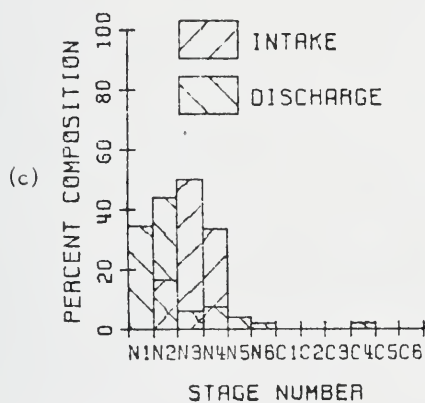
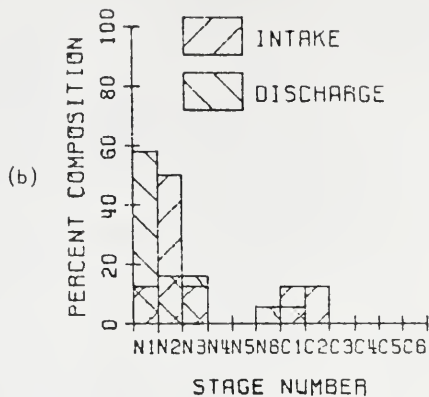
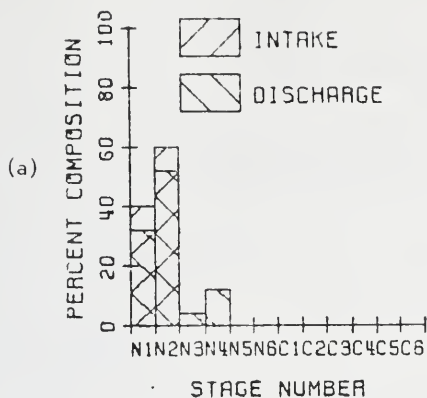
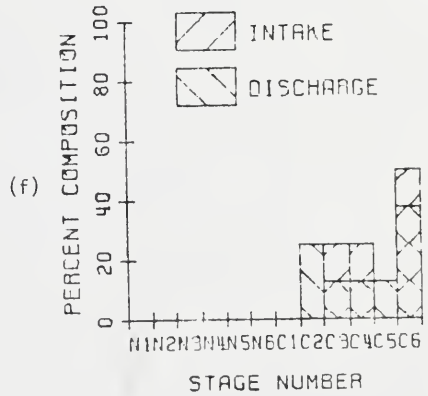
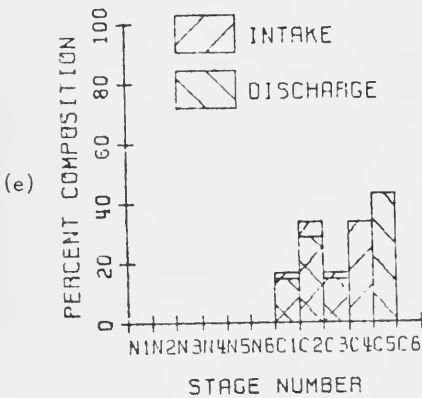
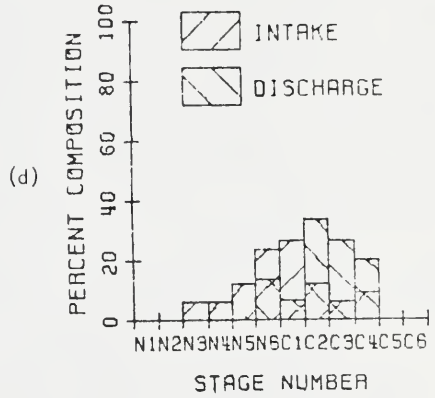
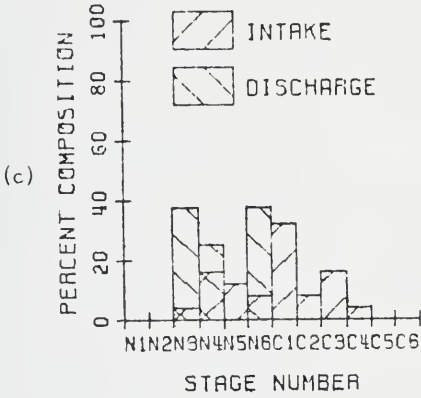
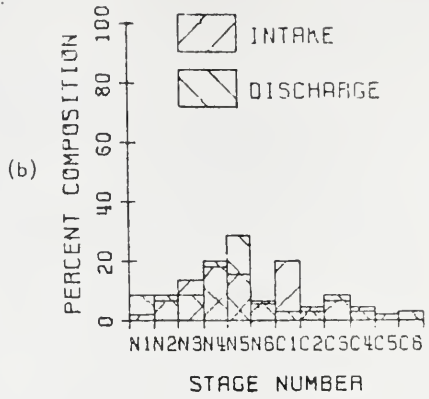
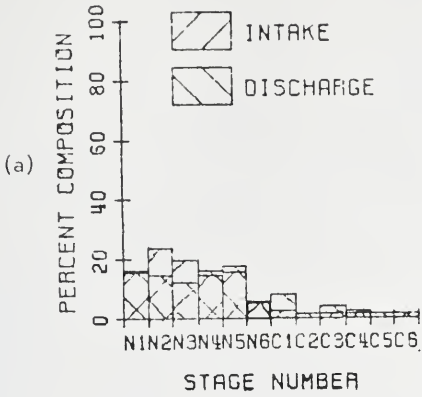


Fig. B-3. Histogram series for Acartia tonsa in growth experiment started on December 26, 1973, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 3, (e) Day 6, (f) Day 8.



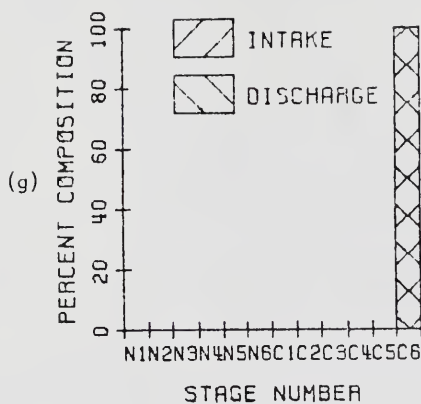
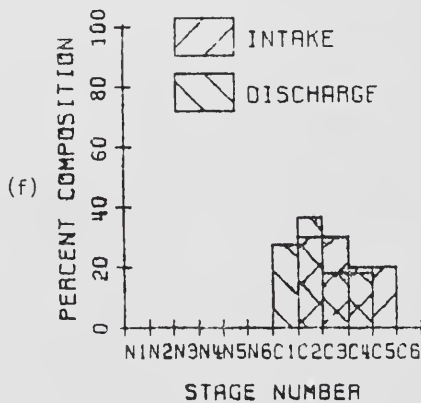
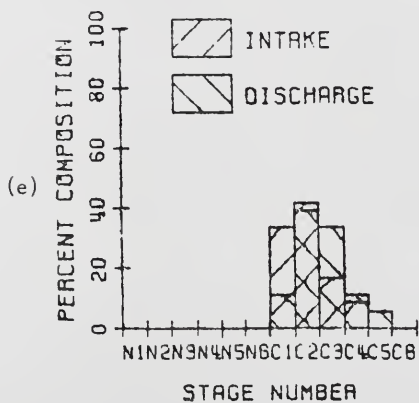
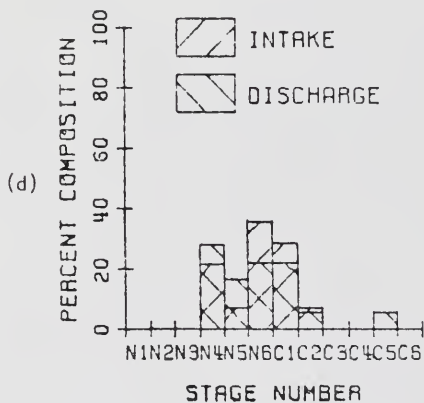
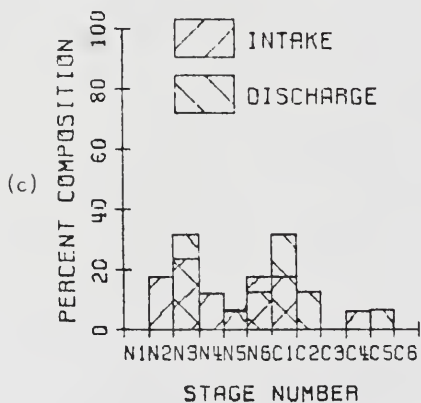
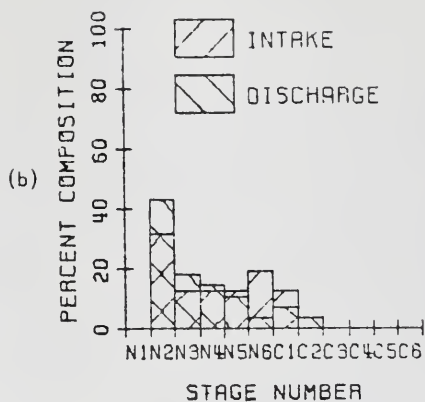
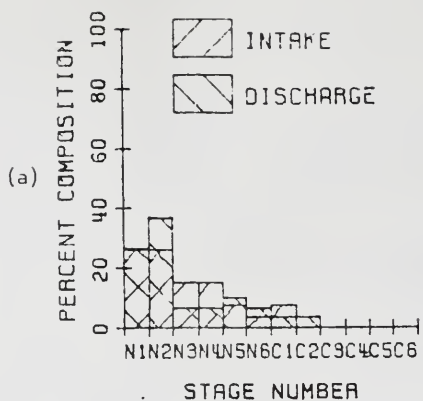


Fig. B-3. Continued
(g) Day 12.

Fig. B-4. Histogram series for Acartia tonsa in growth experiment started on January 22, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 3, (e) Day 6, (f) Day 8.



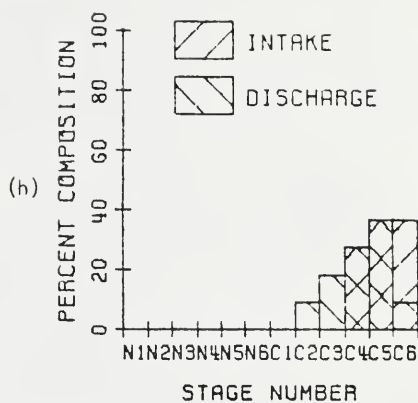
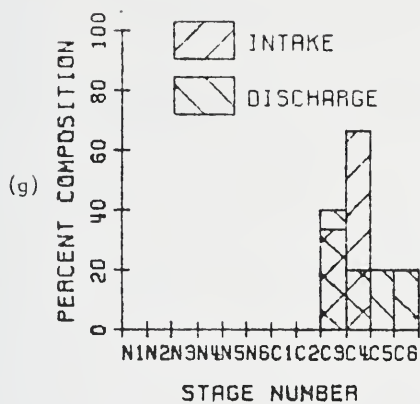
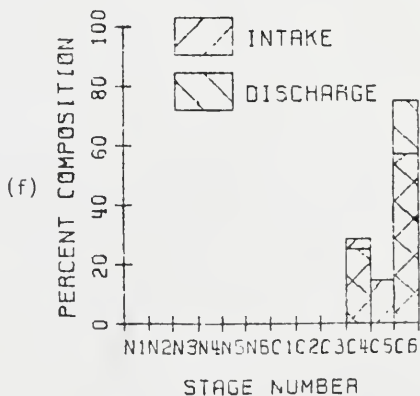
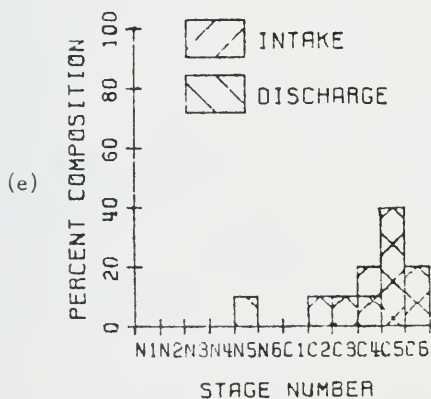
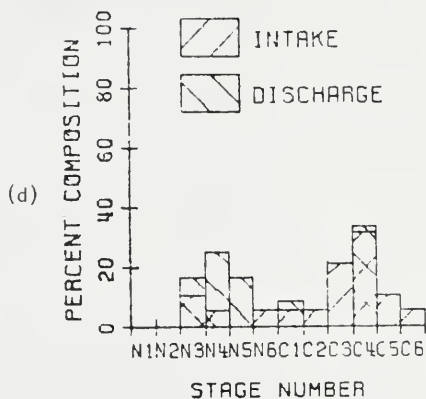
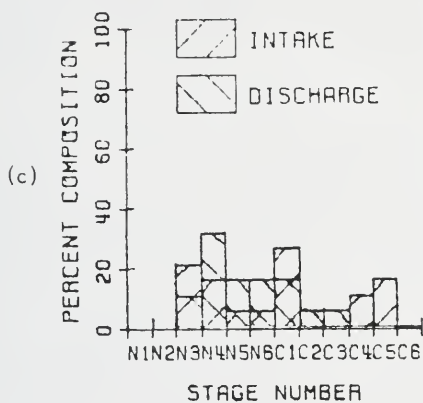
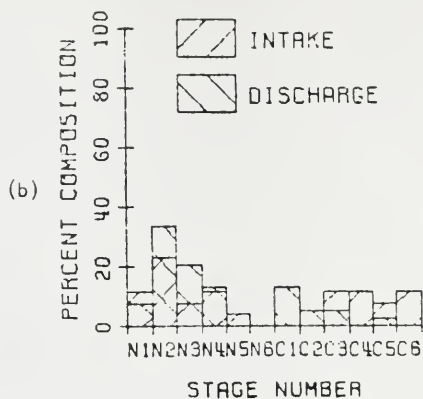
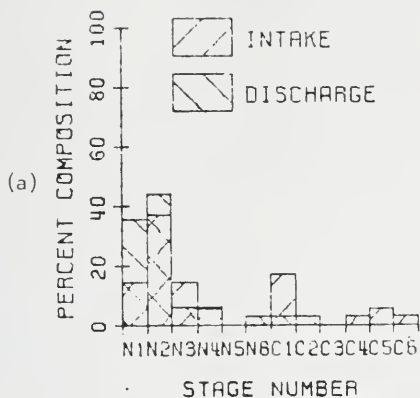


Fig. B-4. Continued
(g) Day 10, (h) Day 12.

Fig. B-5. Histogram series for Acartia tonsa in growth experiment started on February 5, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 3, (e) Day 6, (f) Day 8.



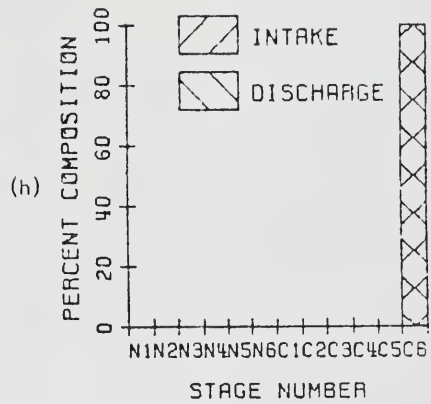
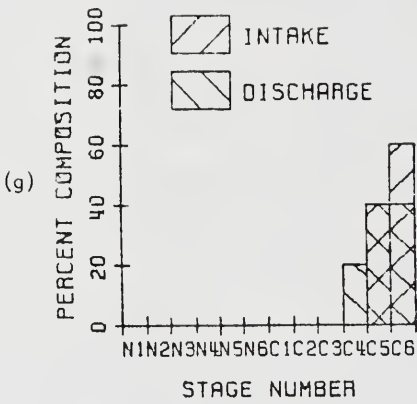


Fig. B-5. Continued
(g) Day 10, (h) Day 11.

Fig. B-6. Histogram series for Acartia tonsa in growth experiment started on March 19, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 3, (e) Day 6, (f) Day 8.

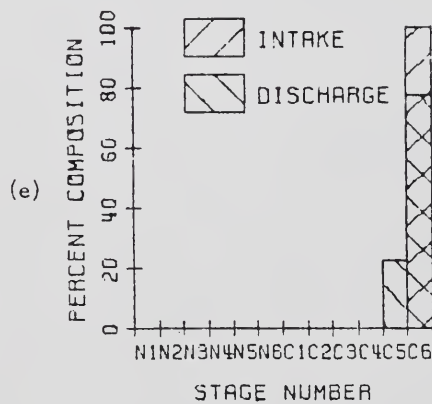
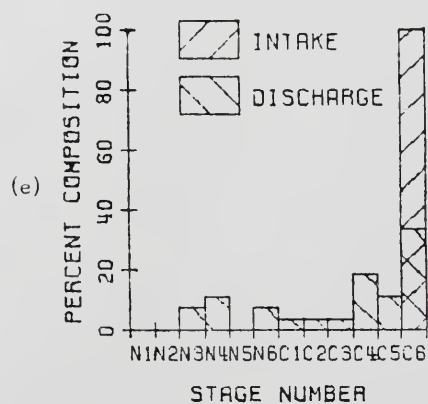
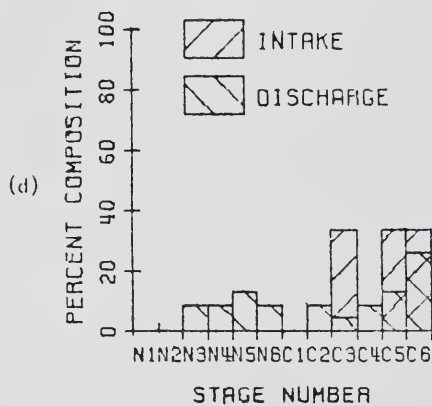
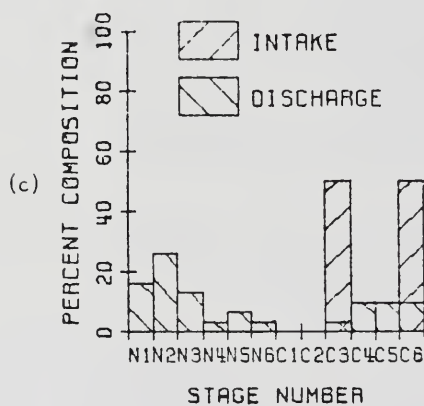
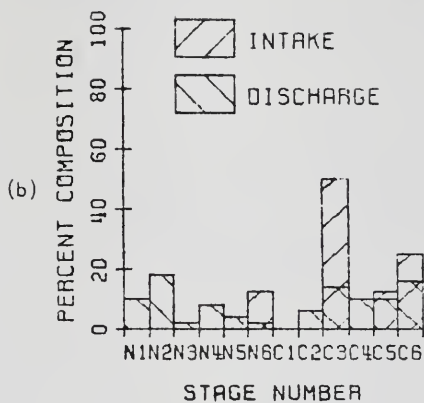
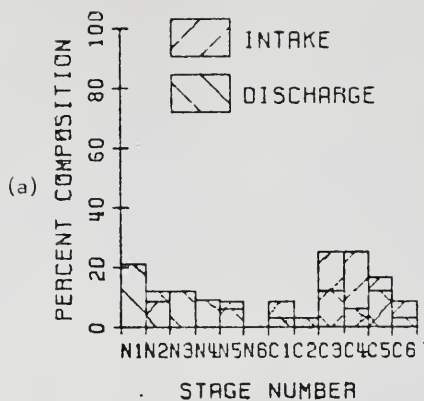
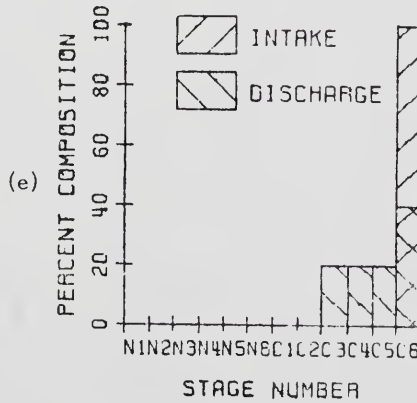
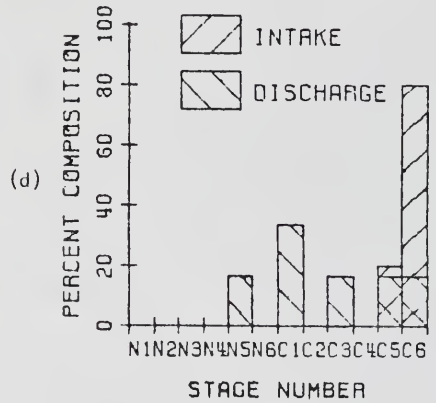
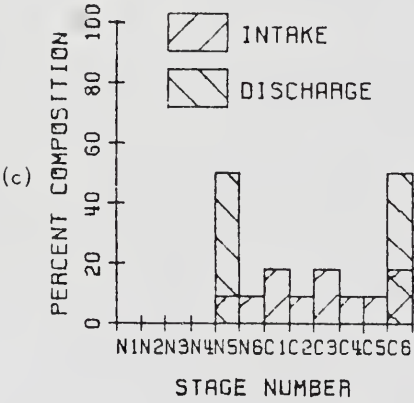
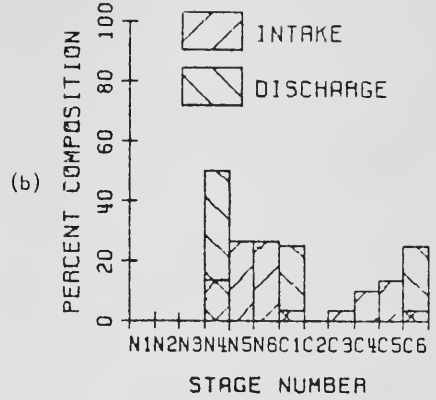
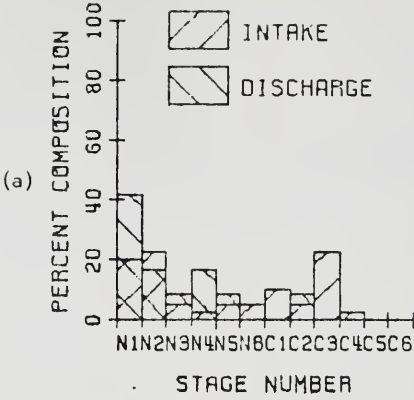


Fig. B-7. Histogram series of Acartia tonsa in growth experiment started on May 28, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 4, (d) Day 6, (e) Day 8.



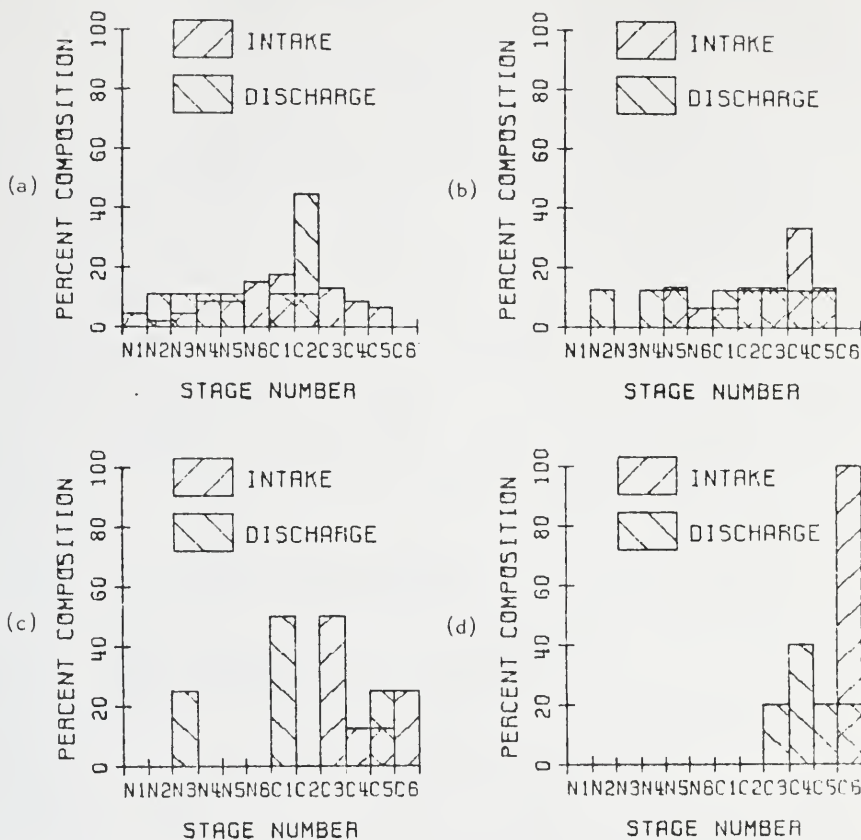
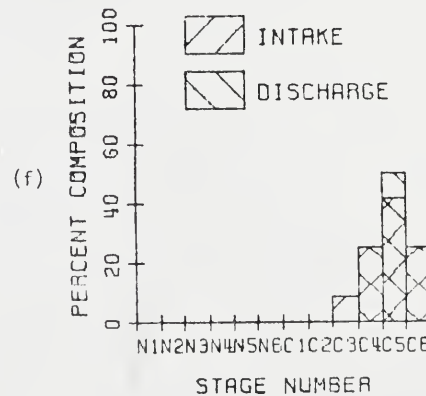
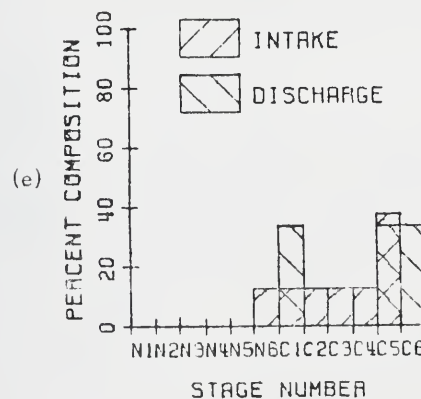
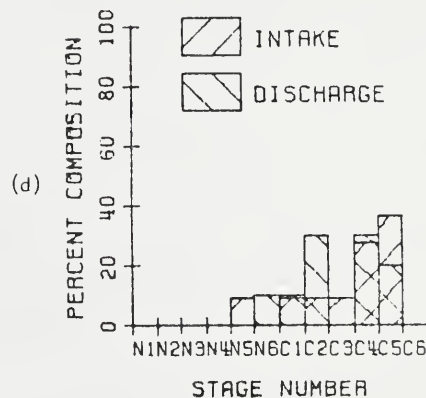
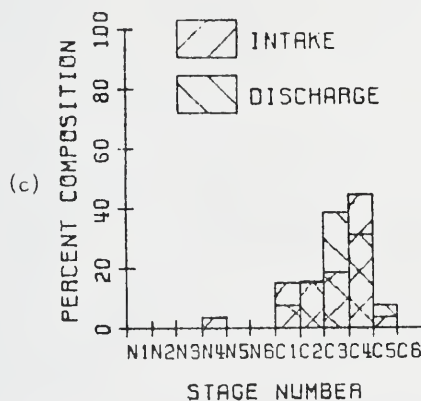
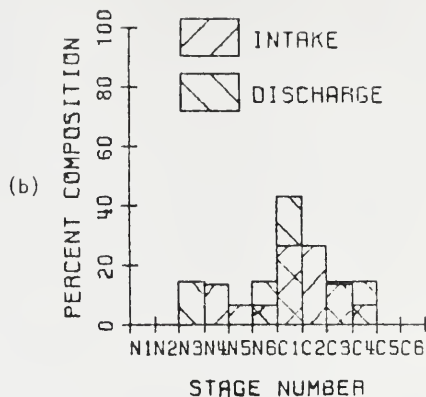
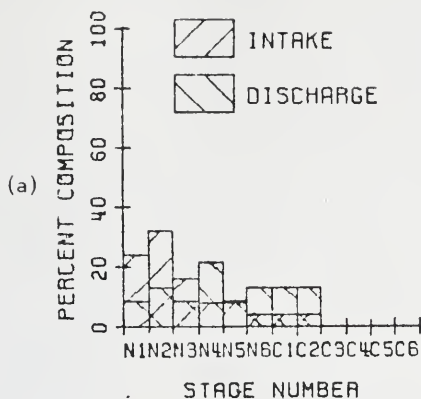


Fig. B-8. Histogram series for *Acartia tonsa* in growth experiment started on June 11, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 6.

Fig. B-9. Histogram series for Acartia tonsa growth experiment started on June 25, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 4, (e) Day 5, (f) Day 6.



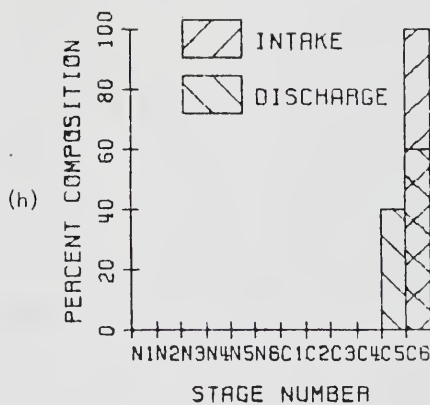
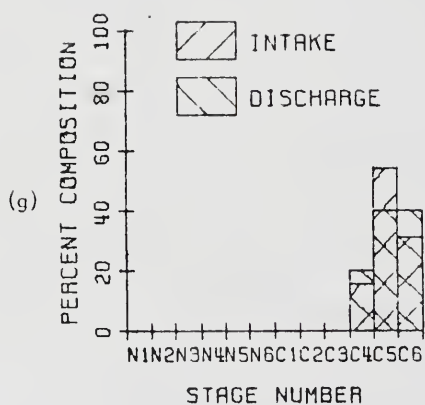
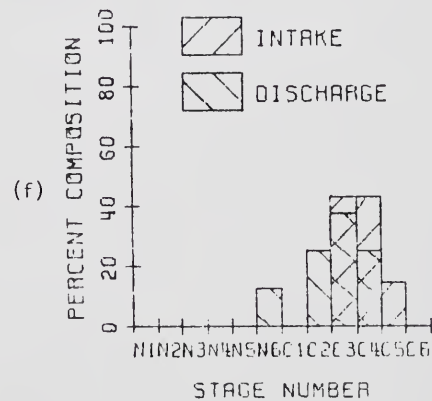
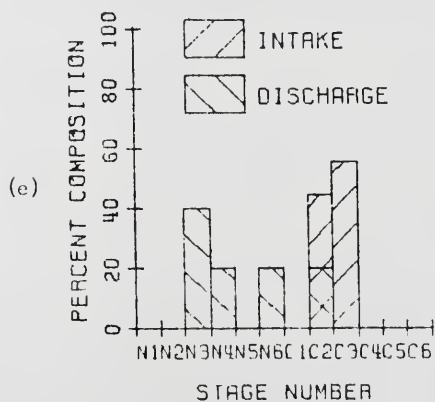
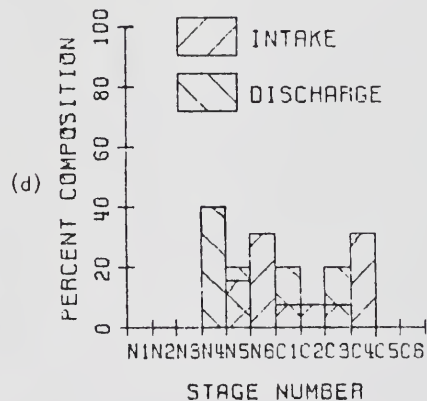
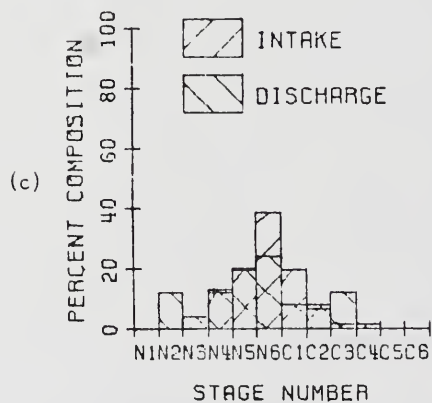
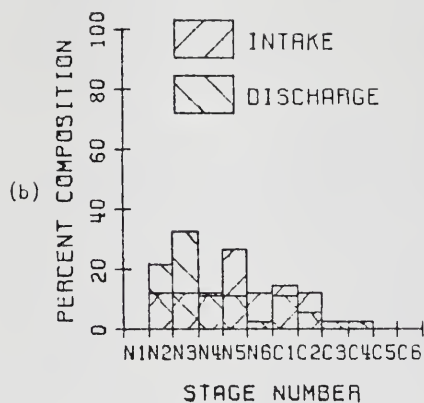
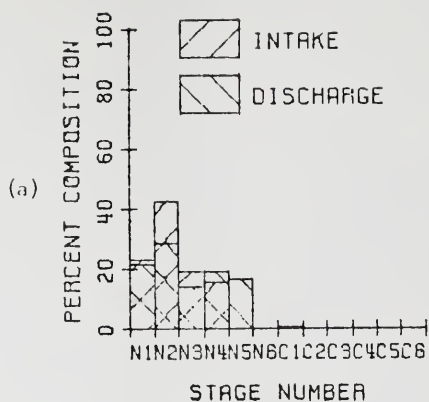


Fig. B-9. Continued
(g) Day 7, (h) Day 8.

Fig. B-10. Histogram series for Acartia tonsa in growth experiment started on July 9, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 3, (c) Day 4, (d) Day 5, (e) Day 6, (f) Day 7.



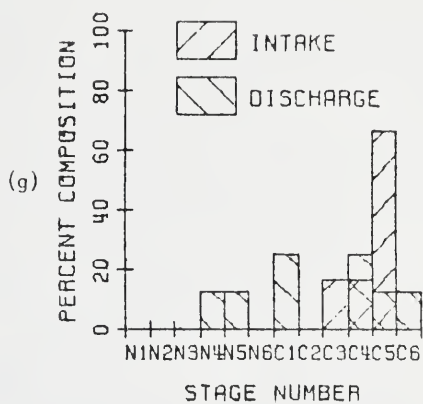
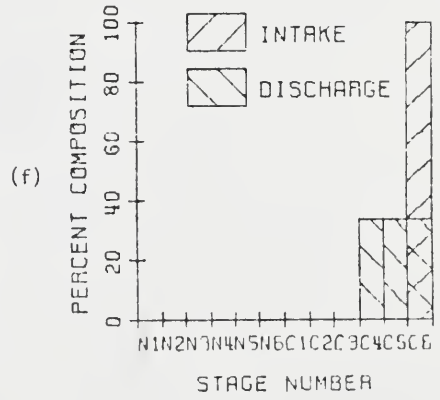
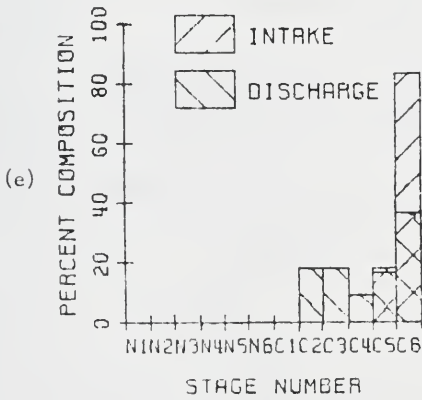
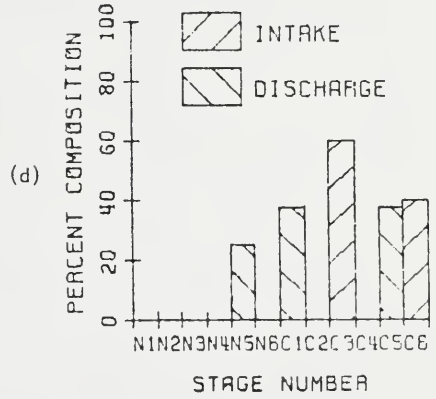
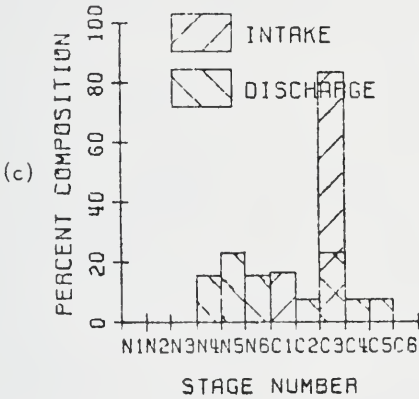
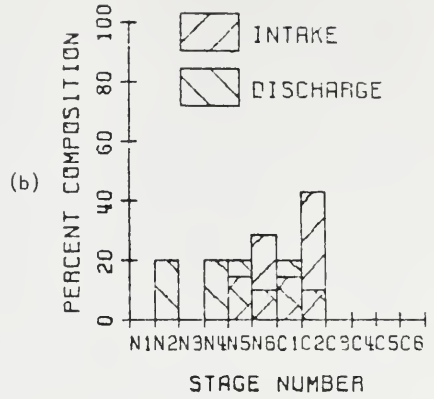
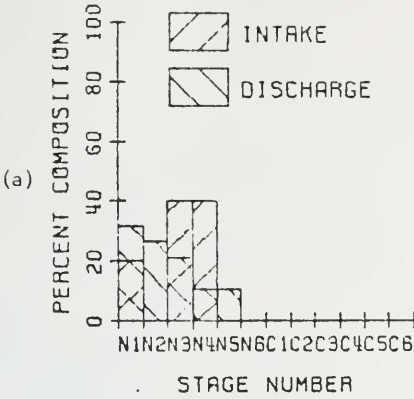


Fig. B-10. Continued
(g) Day 8.

Fig. B-11. Histogram series for Acartia tonsa in growth experiment started on July 23, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 4, (e) Day 5, (f) Day 6.



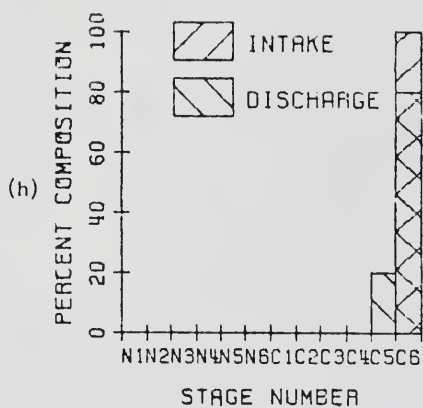
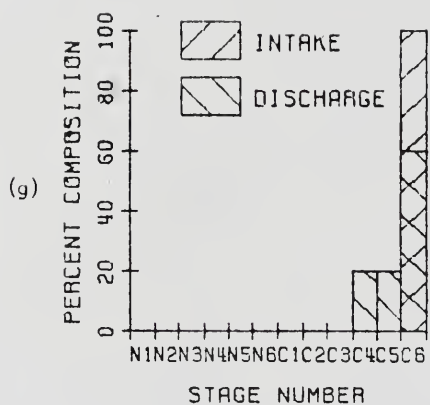
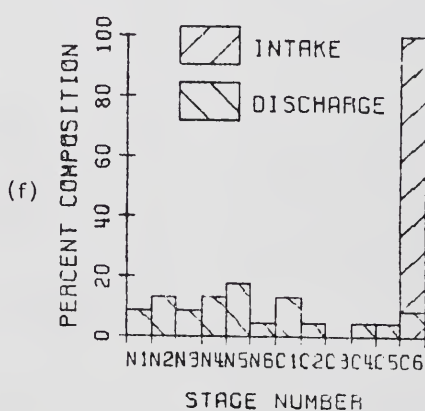
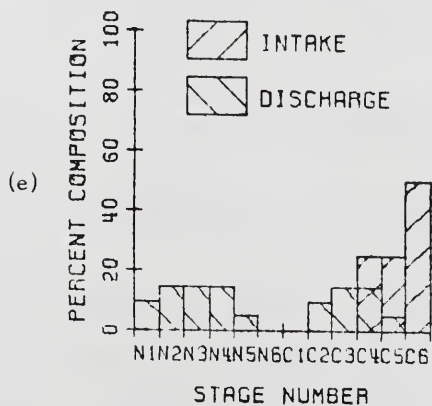
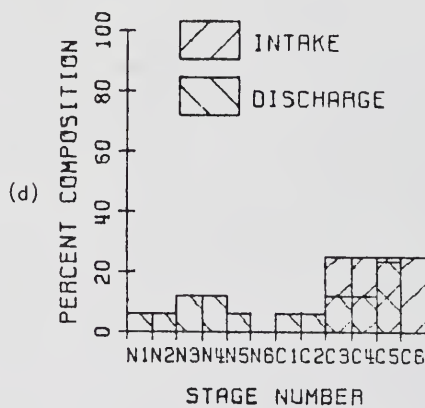
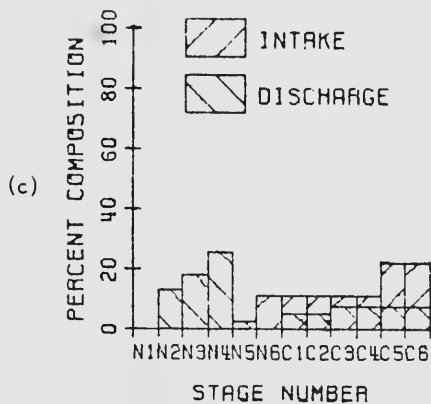
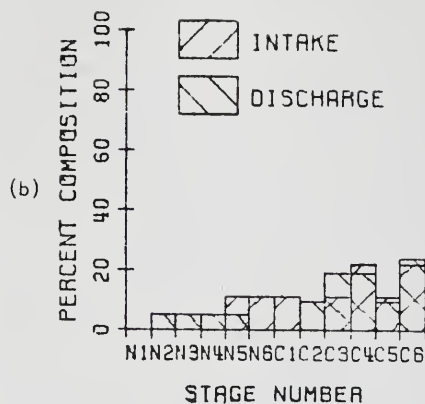
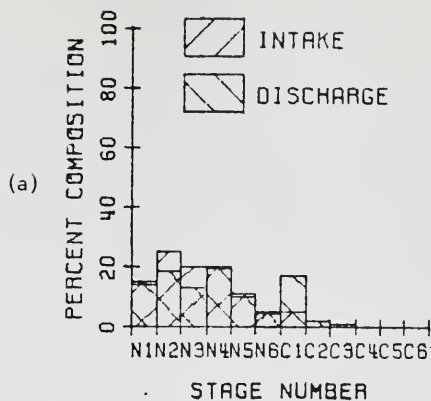


Fig. B-11. Continued
(g) Day 7, (h) Day 8

Fig. B-12. Histogram series for Acartia tonsa in growth experiment started on August 6, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2. (c) Day 3, (d) Day 4, (e) Day 5, (f) Day 6.



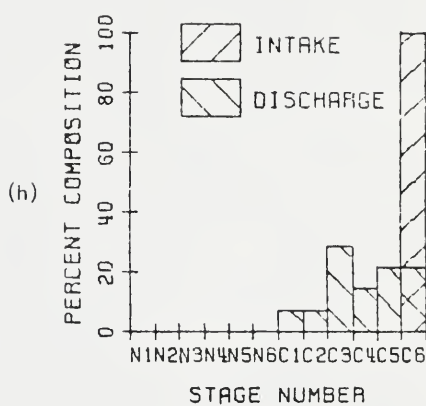
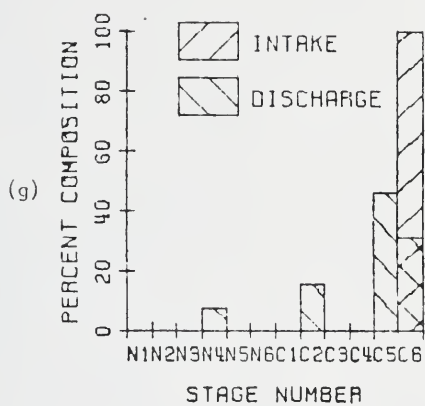
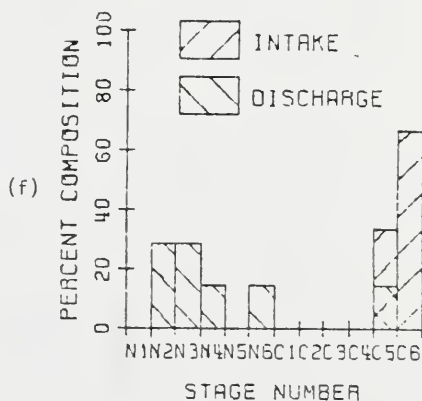
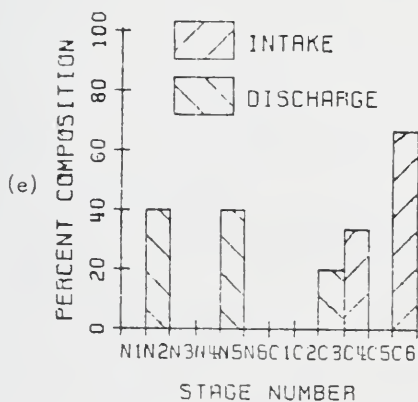
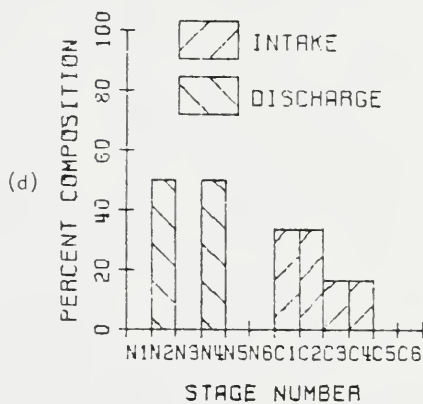
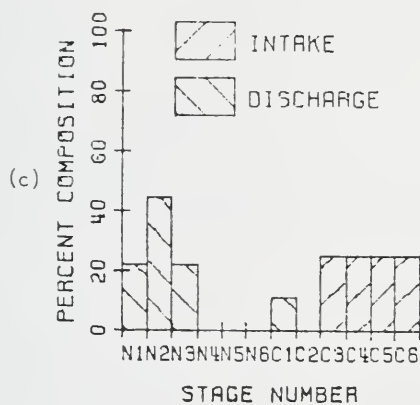
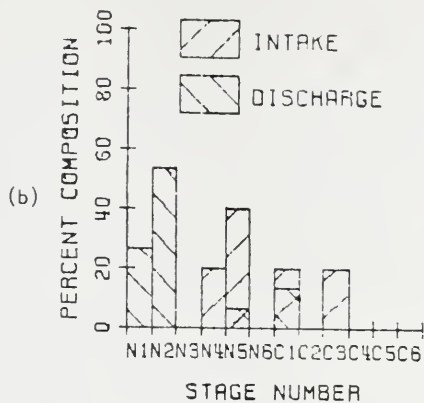
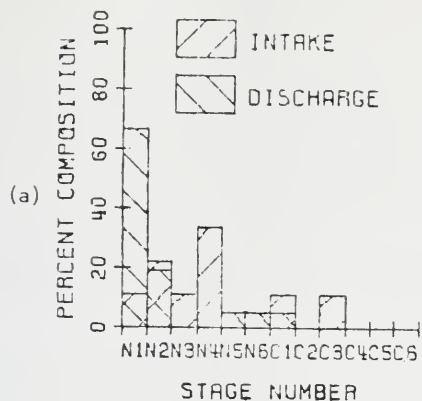


Fig. B-12. Continued
(g) Day 7, (h) Day 8.

Fig. B-13. Histogram series for Acartia tonsa in growth experiment started on August 20, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 4, (e) Day 5, (f) Day 6.



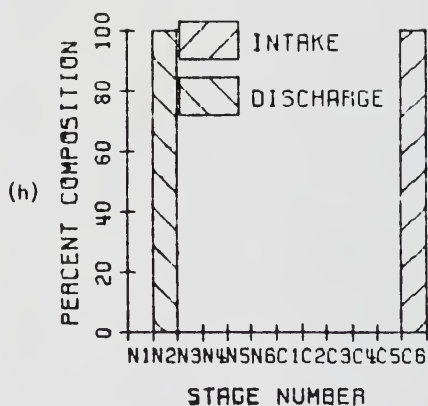
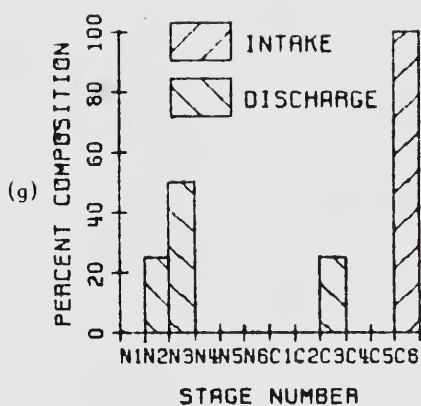
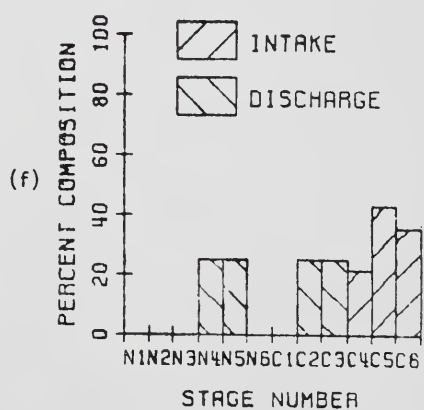
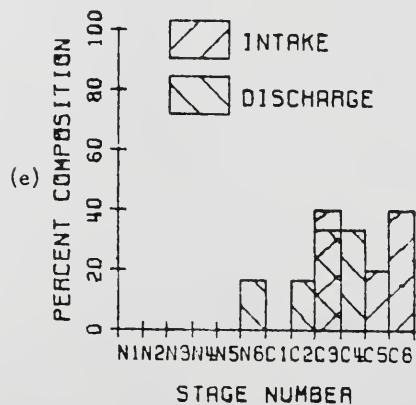
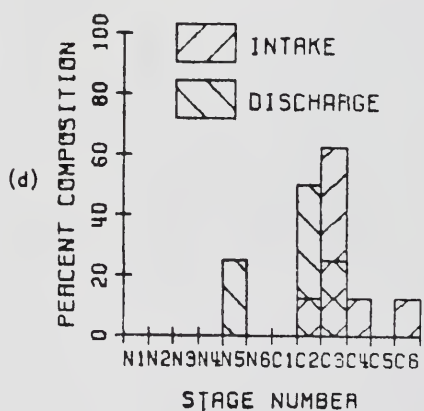
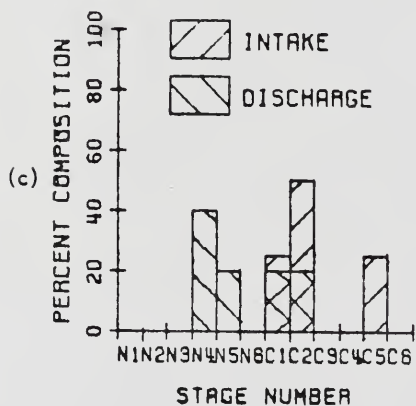
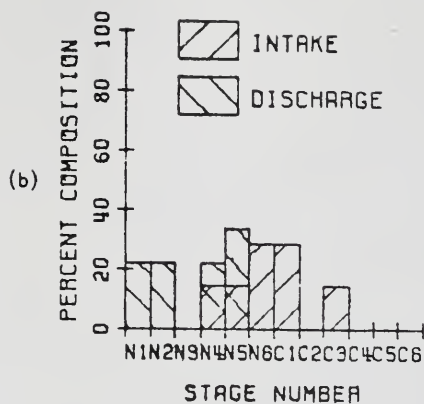
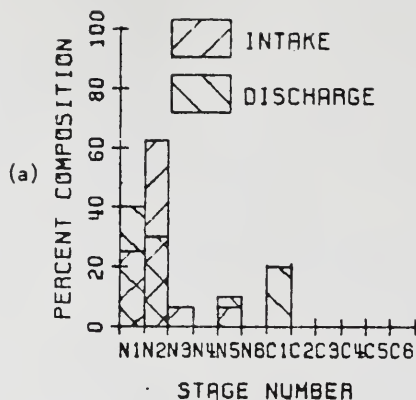


Fig. B-13. Continued
(g) Day 7, (h) Day 8

Fig. B-14. Histogram series for Acartia tonsa in growth experiment started on September 3, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 3, (c) Day 4, (d) Day 5, (e) Day 6, (f) Day 7.



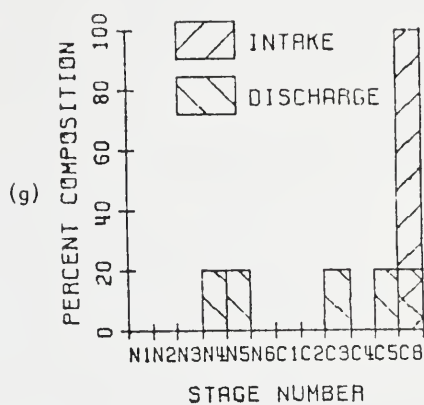
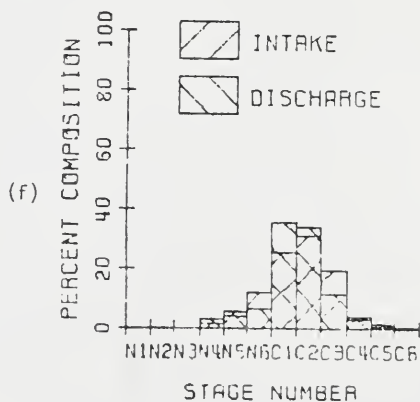
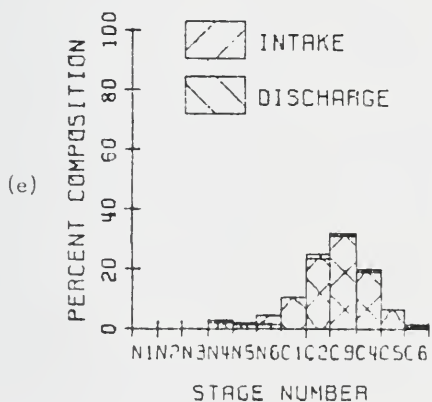
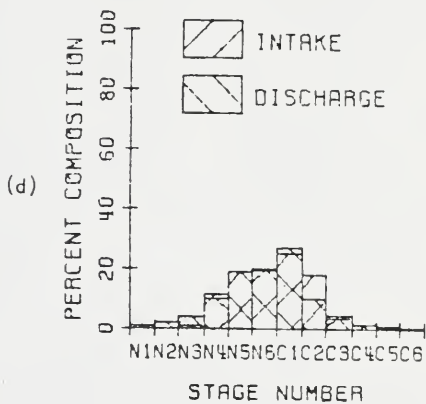
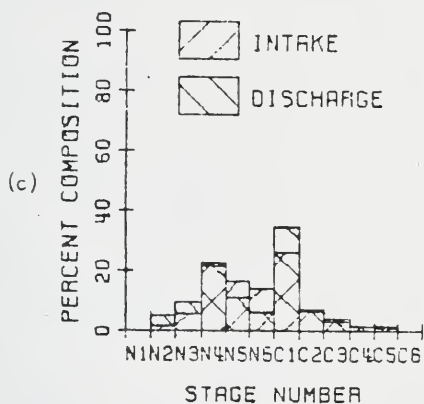
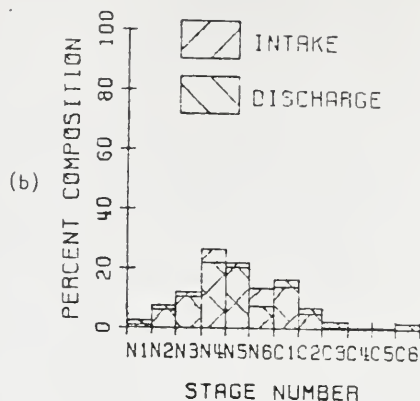
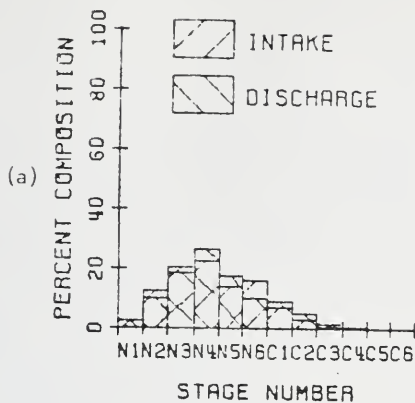


Fig. B-14. Continued
(g) Day 8

Fig. B-15. Histogram series for Oithona brevicornis in growth experiment started on November 27, 1973, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 3, (e) Day 6, (f) Day 9.



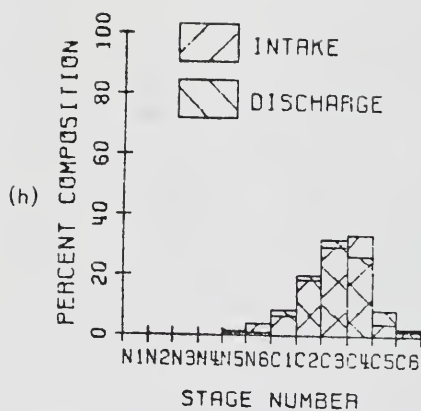
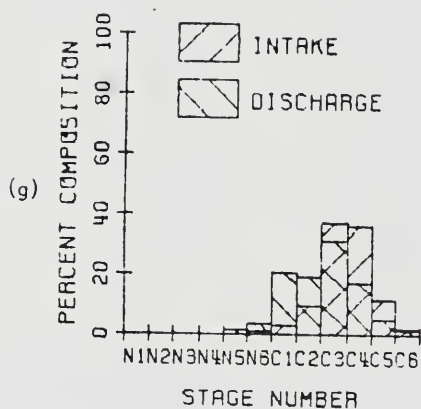


Fig. B-15. Continued
(g) Day 11, (h) Day 13.8.

Fig. B-16. Histogram series for Oithona brevicornis in growth experiment started on December 26, 1973, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 3, (e) Day 6, (f) Day 8.

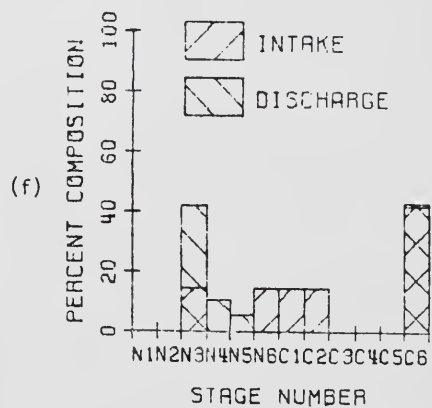
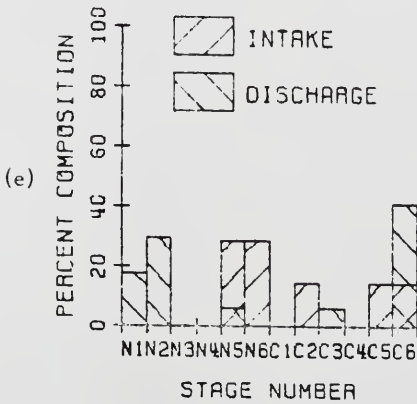
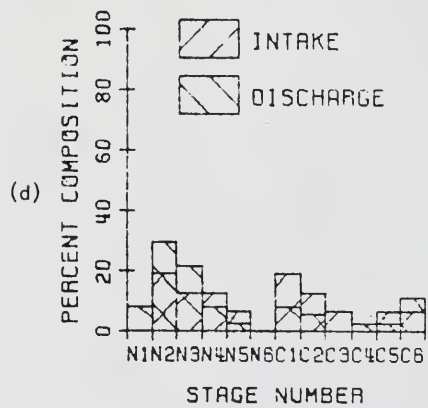
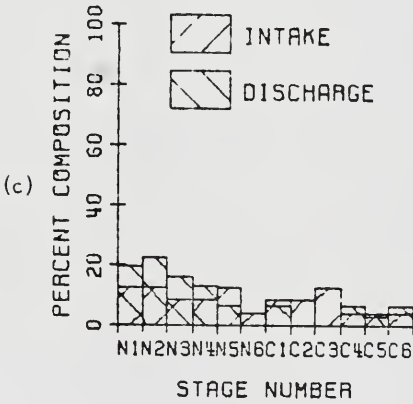
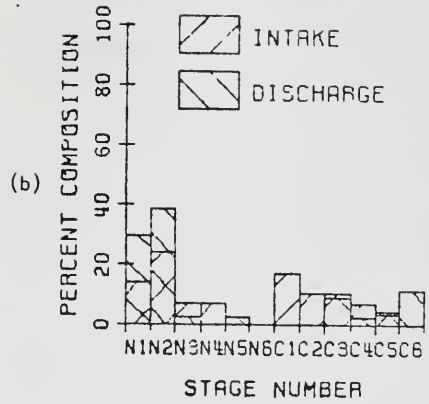
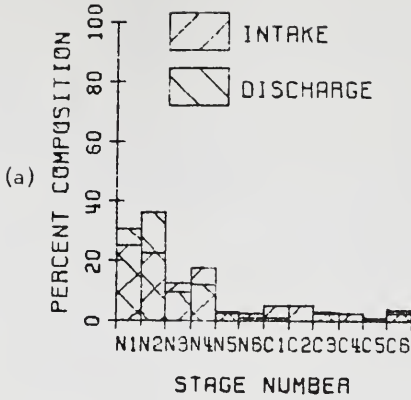
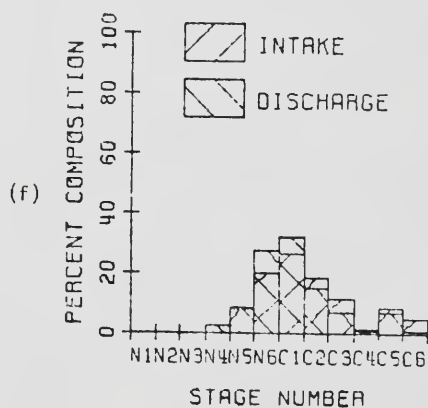
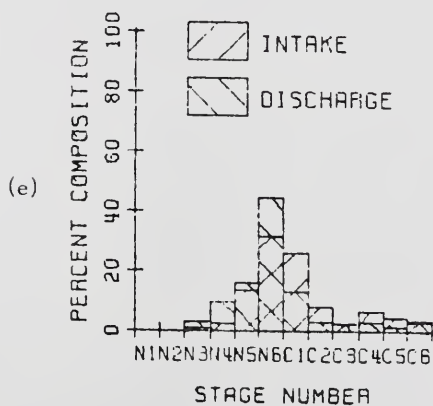
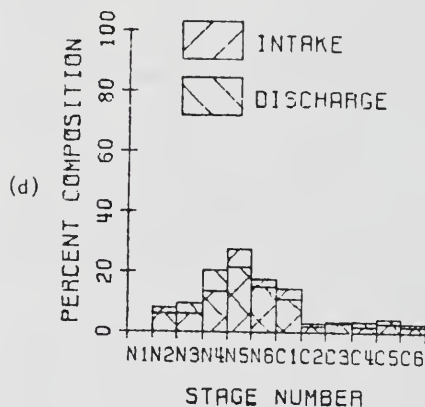
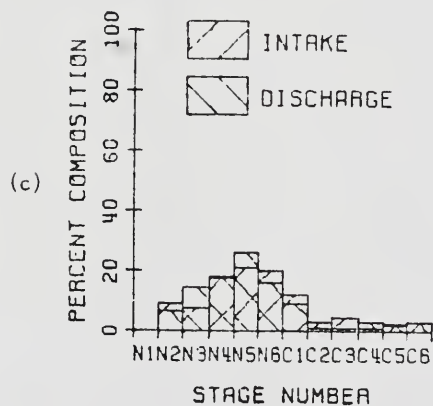
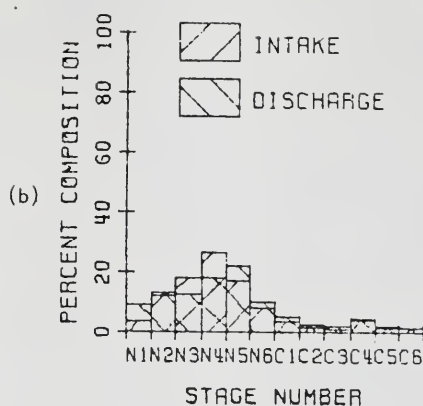
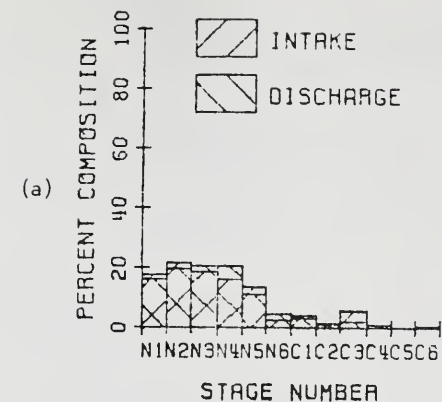


Fig. B-17. Histogram series for Oithona brevicornis in growth experiment started on January 22, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 3, (e) Day 6, (f) Day 8.



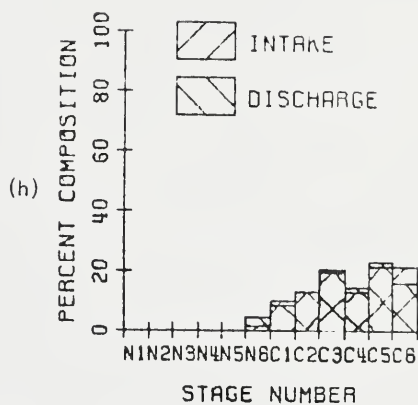
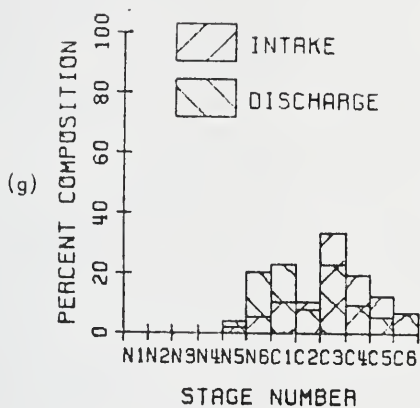


Fig. B-17. Continued
(g) Day 10, (h) Day 12.

Fig. B-18. Histogram series for Oithona brevicornis in growth experiment started on February 5, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 3, (e) Day 6.

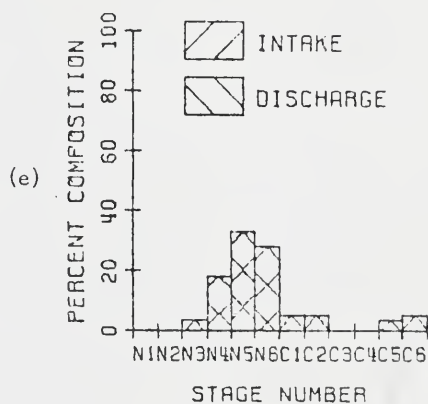
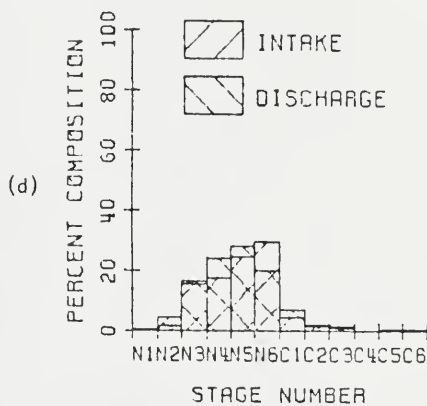
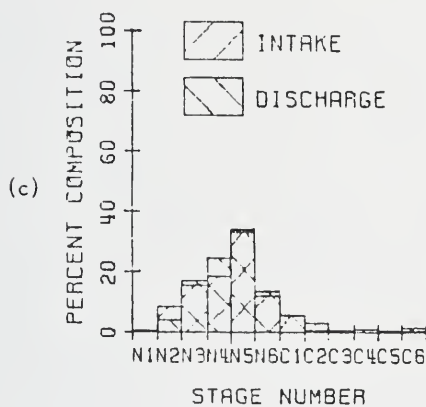
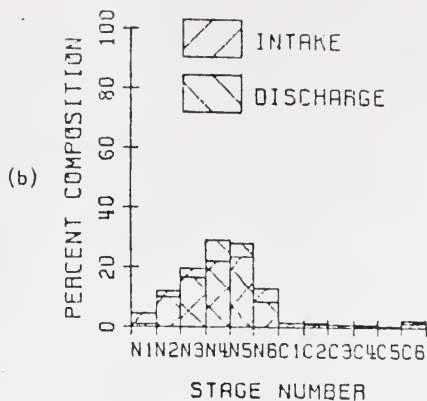
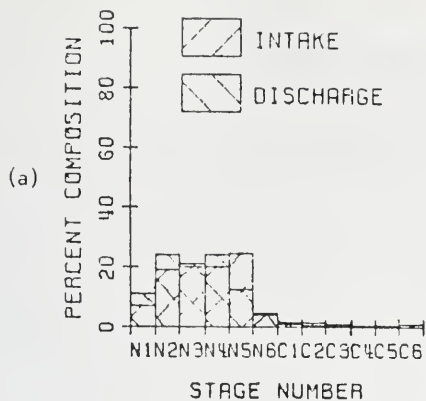
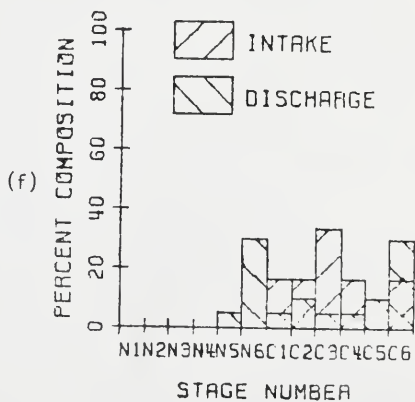
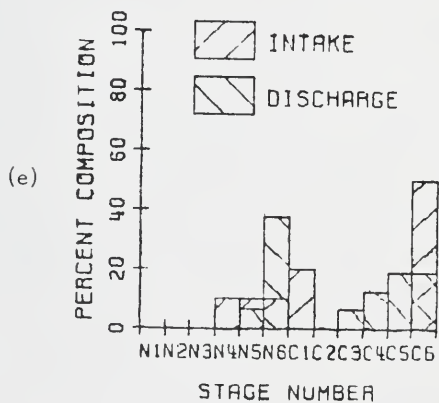
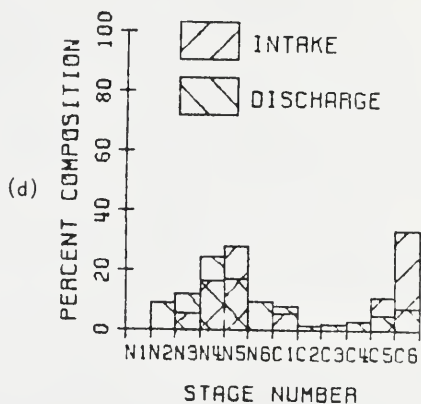
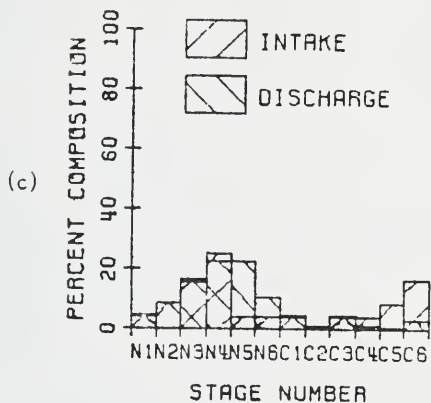
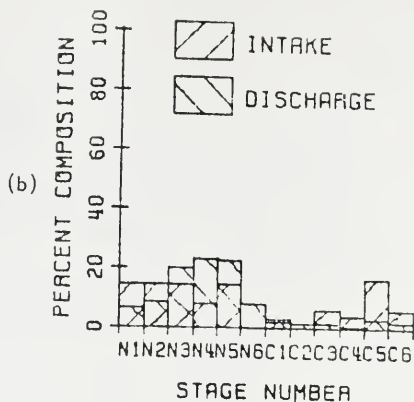
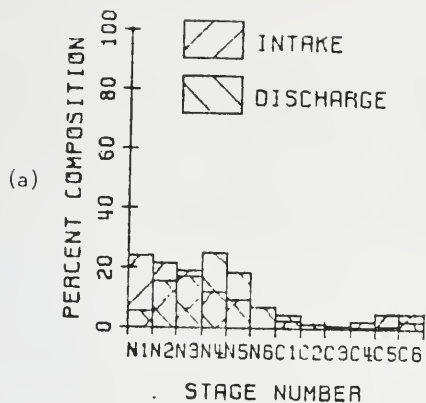


Fig. B-19. Histogram series for Oithona brevicornis in growth experiment started on March 19, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 3, (e) Day 6, (f) Day 8.



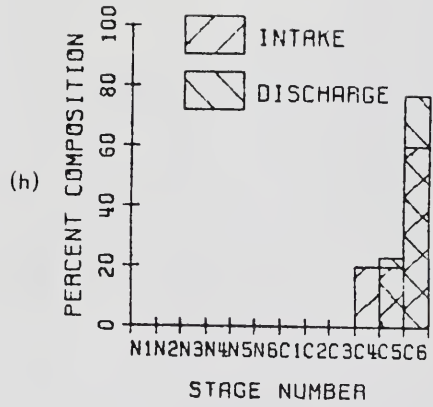
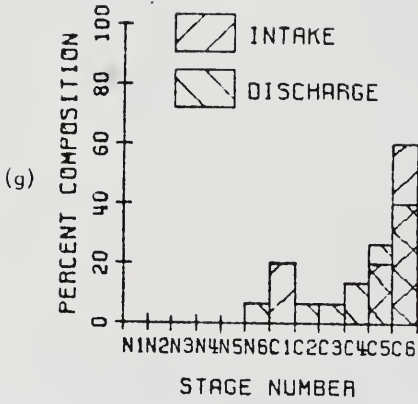
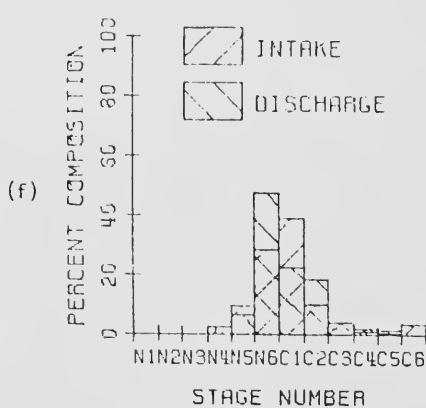
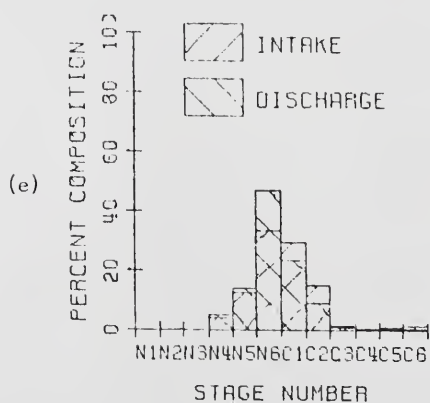
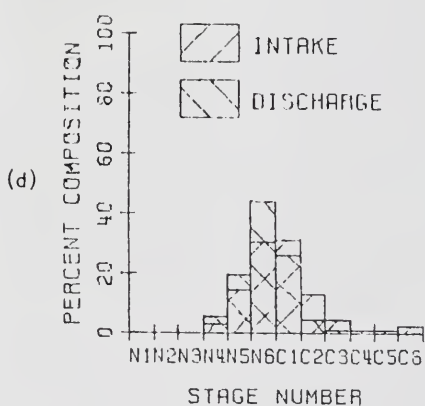
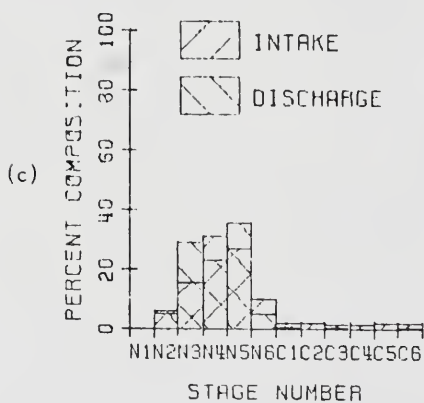
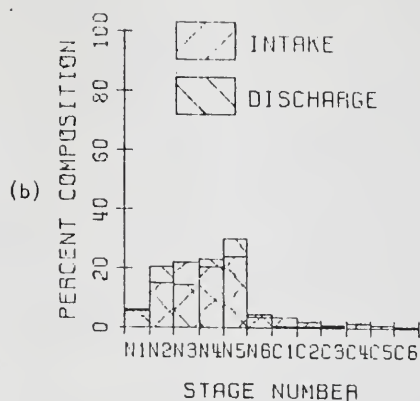
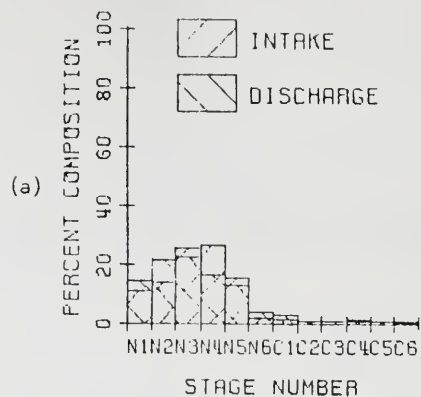


Fig. B-19. Continued
(g) Day 10, (h) Day 13.

Fig. B-20. Histogram series for Oithona brevicornis in growth experiment started on April 24, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 1, (c) Day 2, (d) Day 5, (e) Day 6, (f) Day 7.



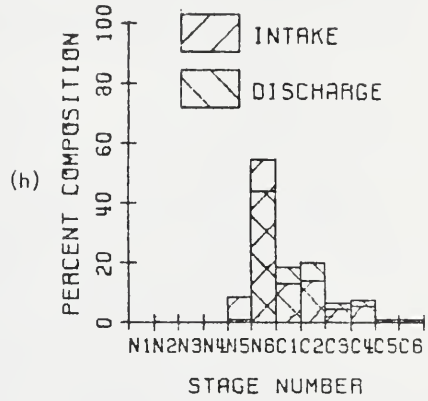
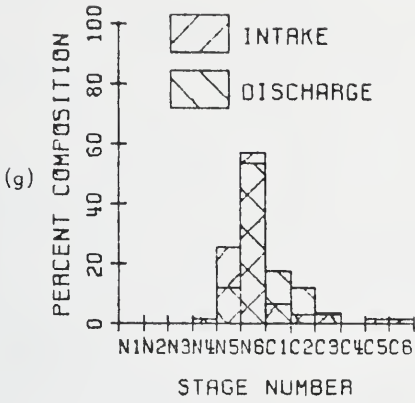
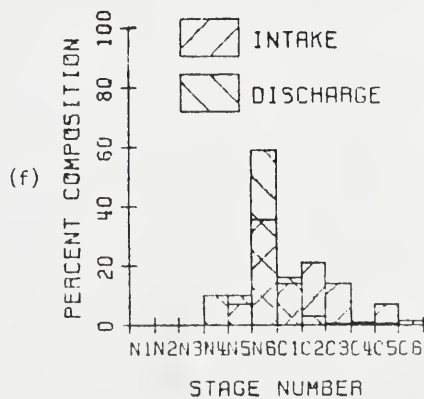
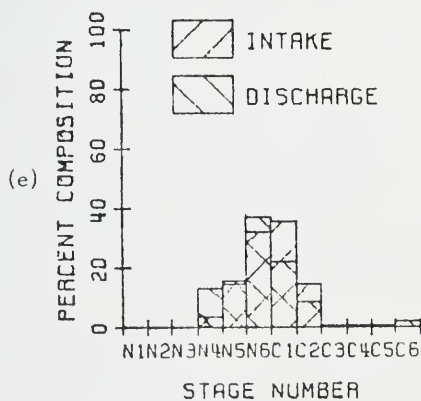
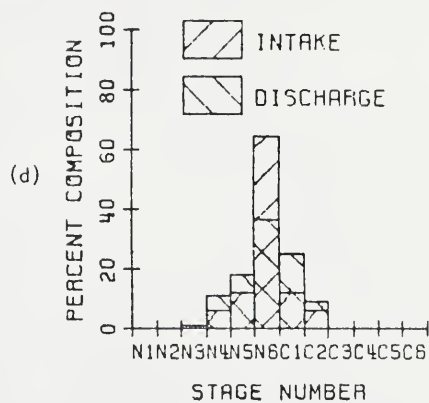
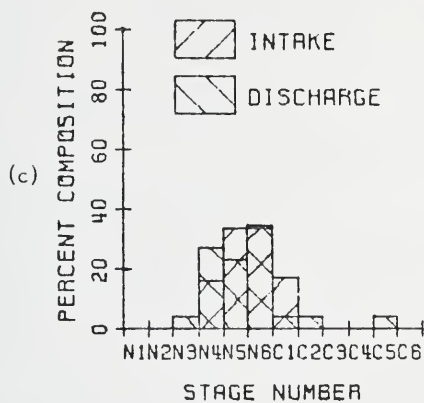
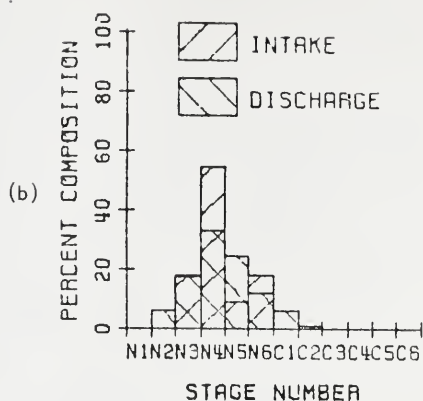
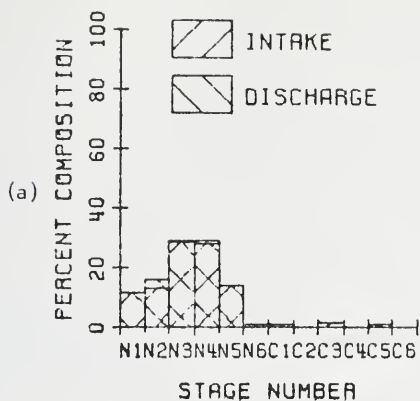


Fig. B-20. Continued
(g) Day 8, (h) Day 9.

Fig. B-21. Histogram series for Oithona brevicornis in growth experiment started on May 14, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 3, (c) Day 6, (d) Day 7, (e) Day 8, (f) Day 9.3.



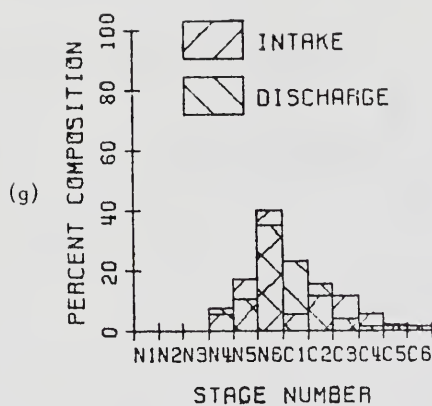
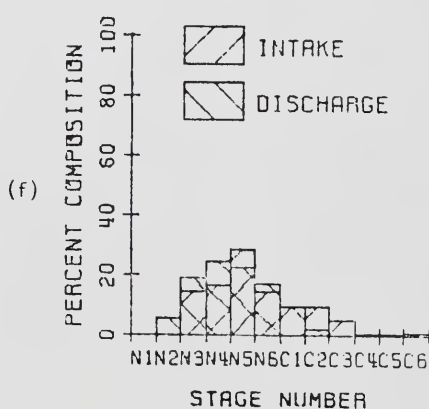
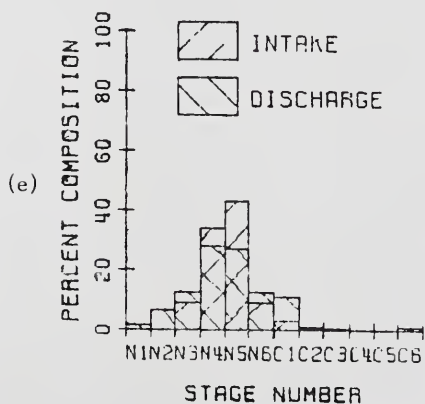
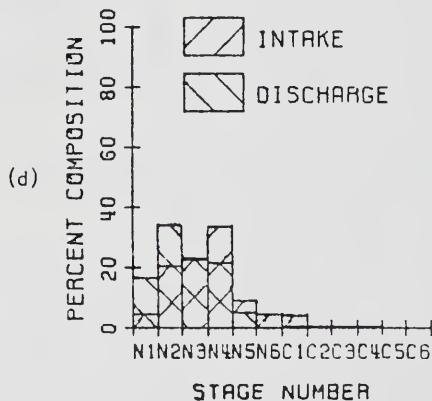
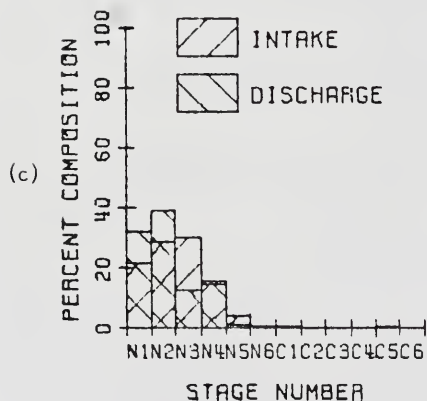
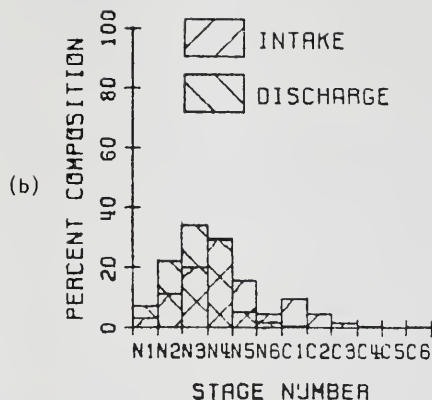
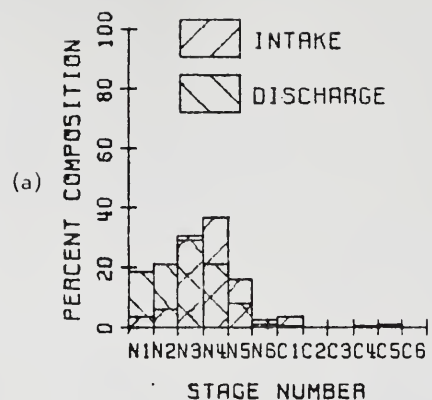


Fig. B-21. Continued
(g) Day 12

Fig. B-22. Histogram series for Oithona brevicornis in growth experiment started on May 28, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 4, (e) Day 6, (f) Day 7.



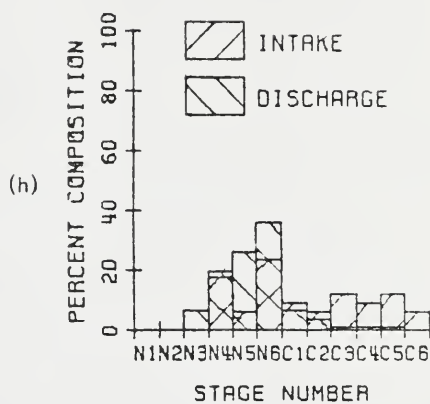
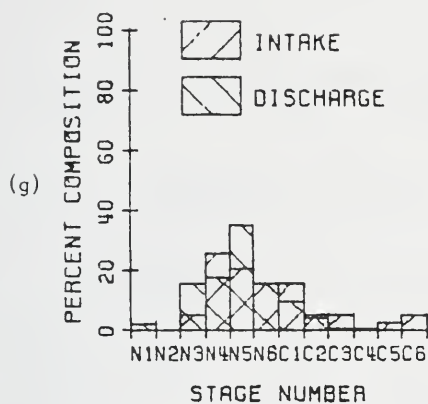


Fig. B-22. Continued
(g) Day 8, (h) Day 9.

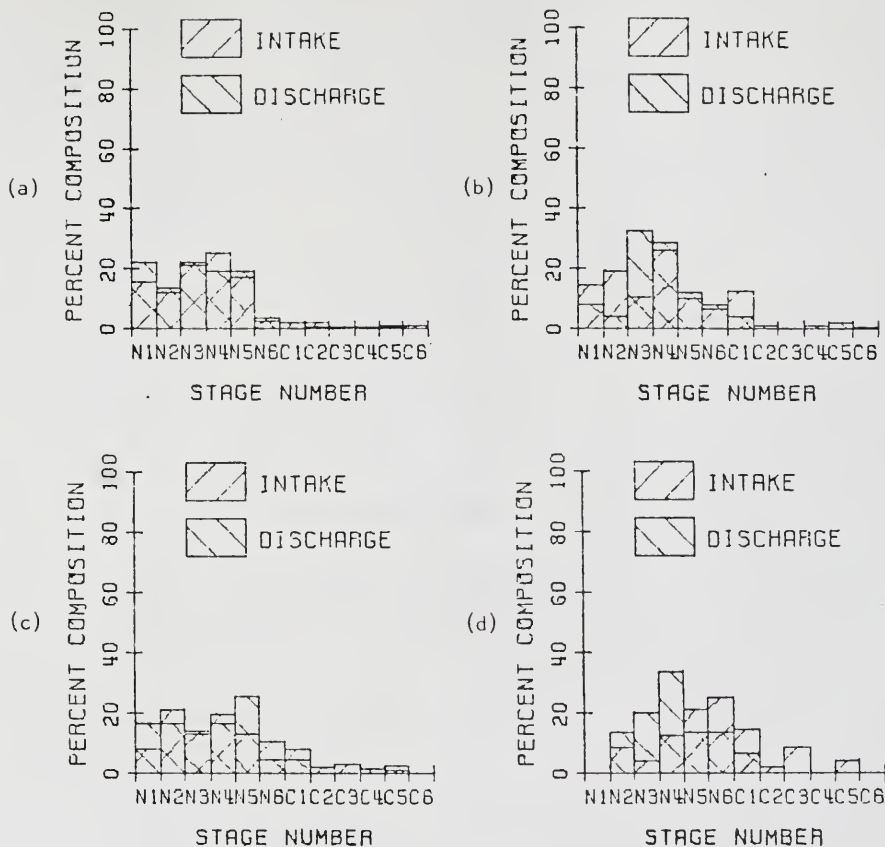
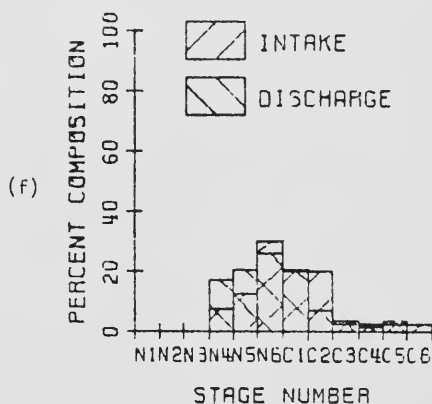
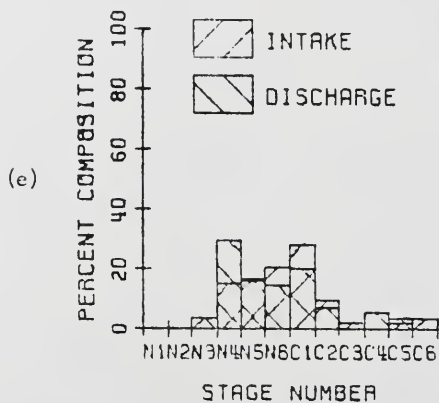
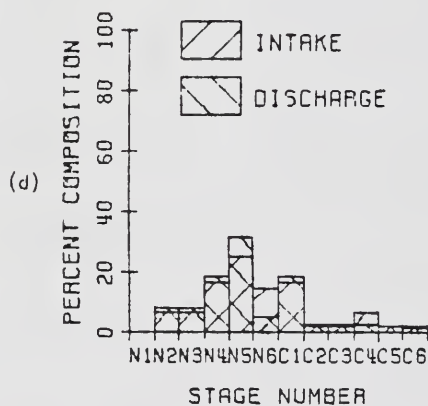
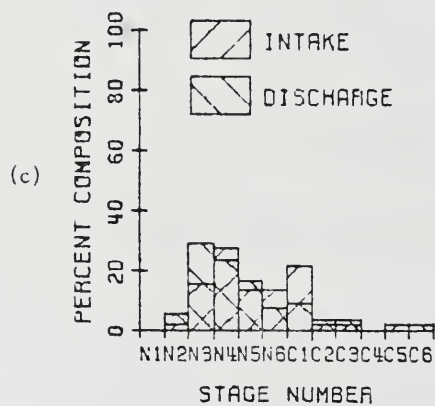
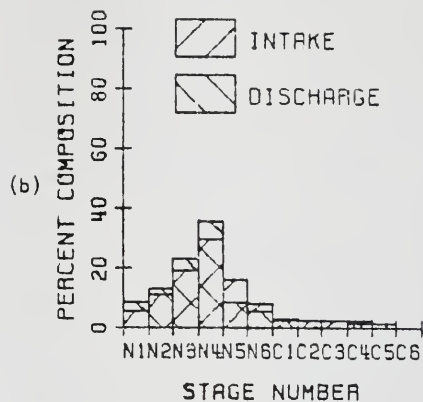
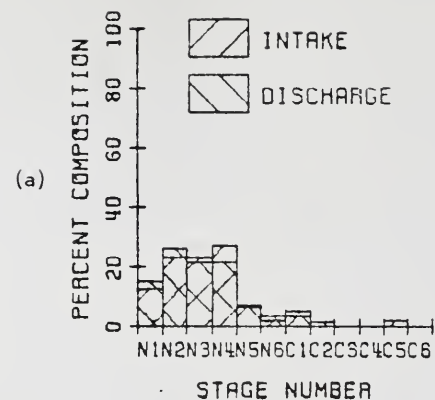


Fig. B-23. Histogram series for *Oithona brevicornis* in growth experiment started on June 11, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 6.

Fig. B-24. Histogram series for Oithona brevicornis in growth experiment started on June 25, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 4, (e) Day 5, (f) Day 6.



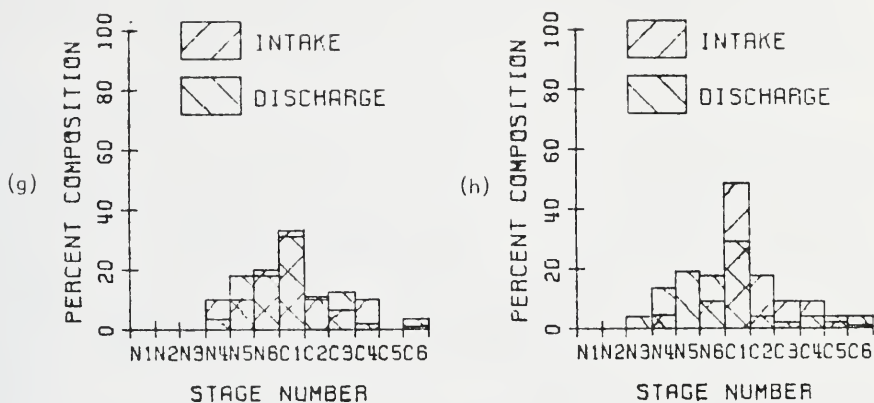
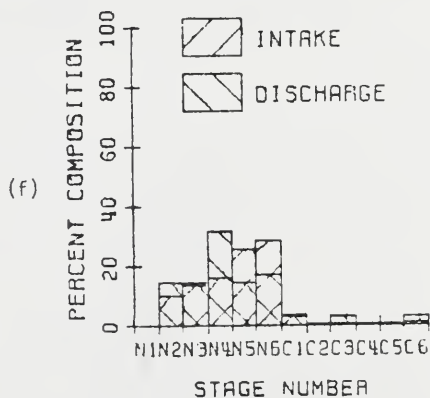
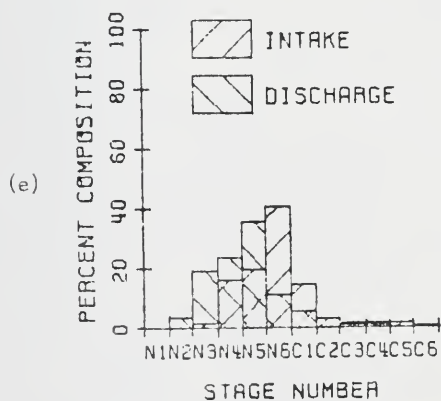
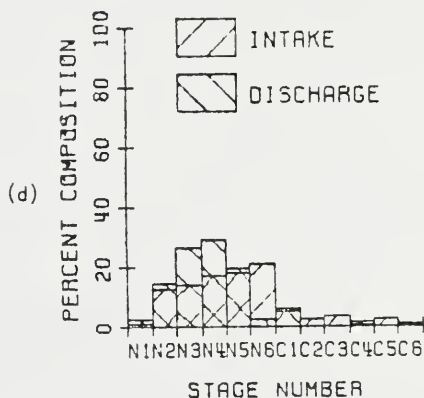
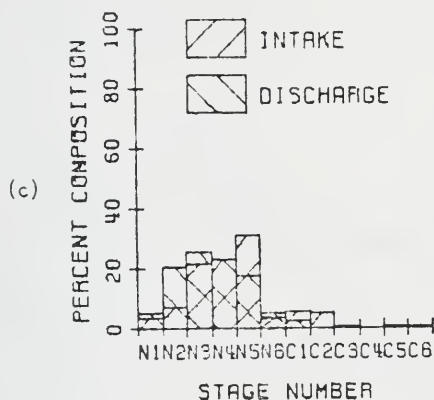
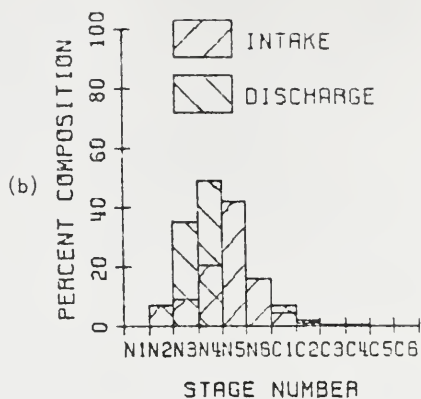
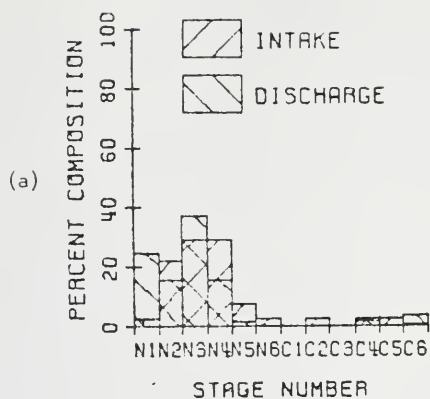


Fig. B-24. Continued
(g) Day 7, (h) Day 8.

Fig. 8-25. Histogram series for Oithona brevicornis in growth experiment started on July 9, 1974, showing changes in population structures with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 4, (e) Day 5, (f) Day 6.



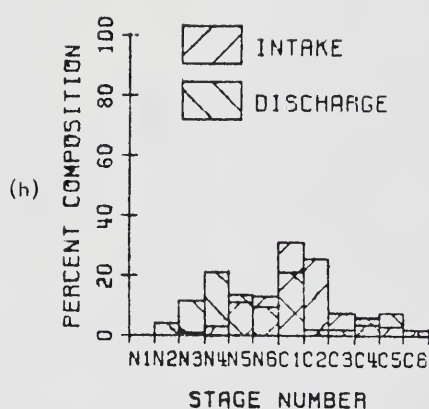
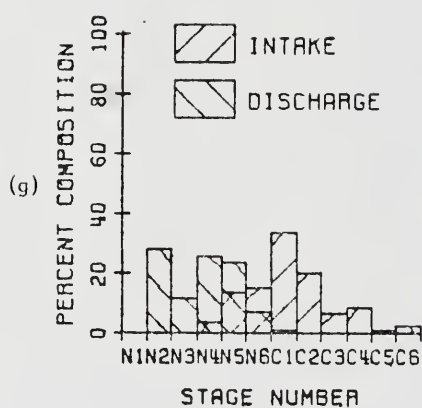
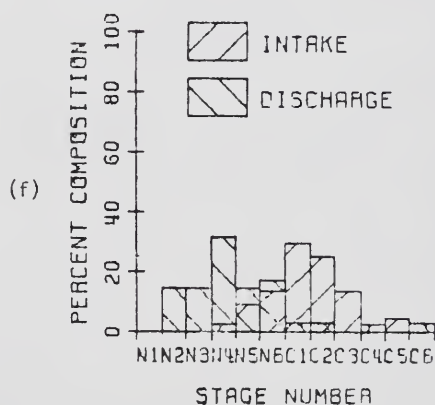
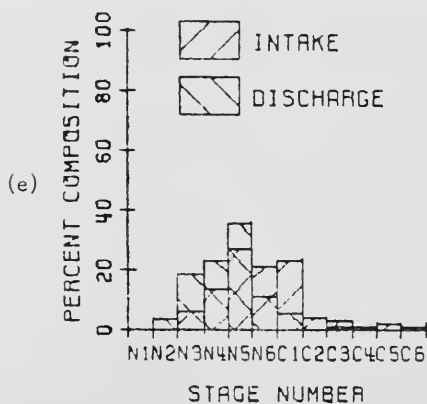
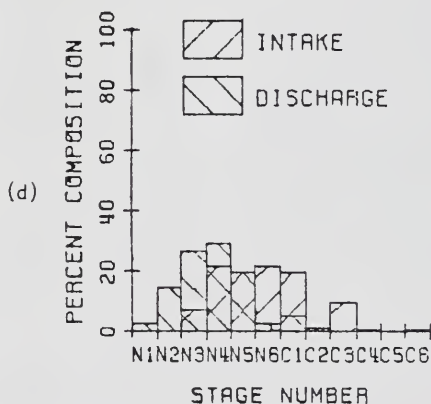
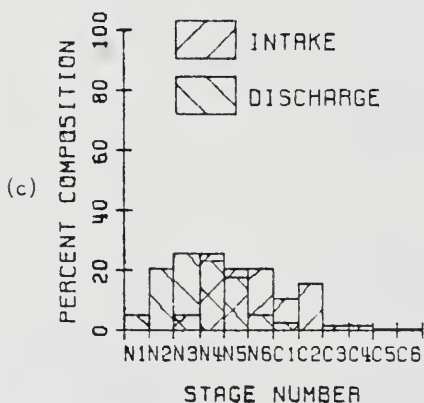
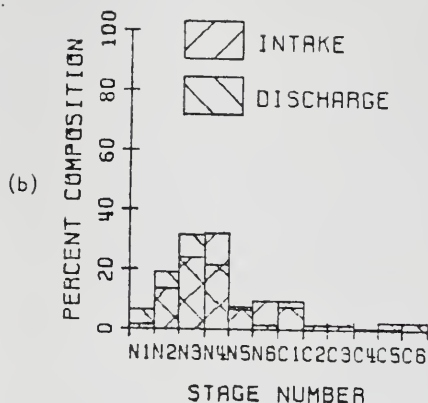
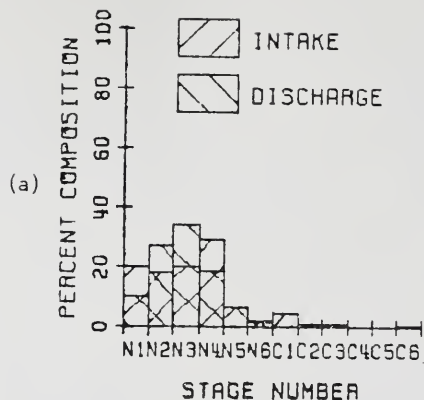


Fig. B-25. Continued
 (g) Day 7, (h) Day 8.

Fig. B-26. Histogram series for Oithona brevicornis in growth experiment started on July 23, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 4, (e) Day 5, (f) Day 6.



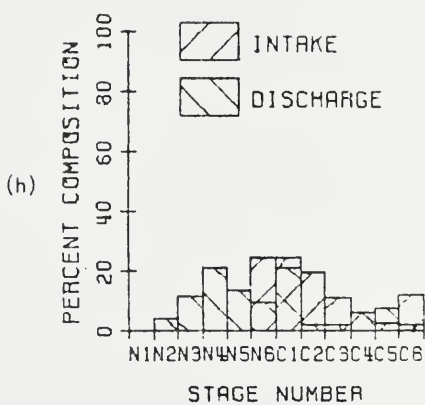
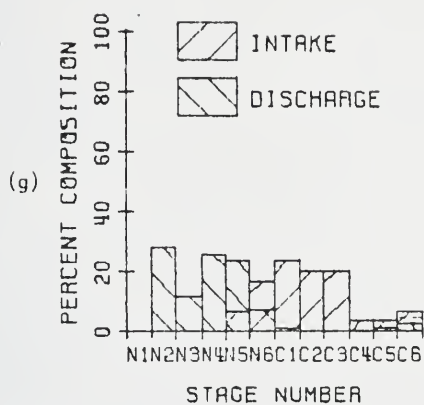
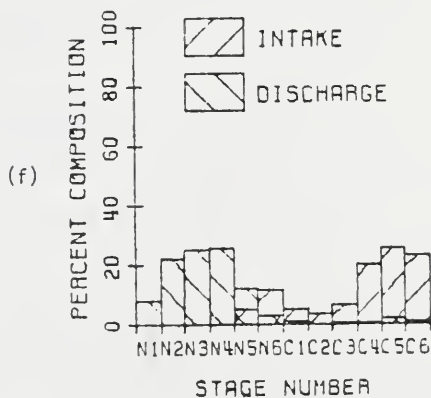
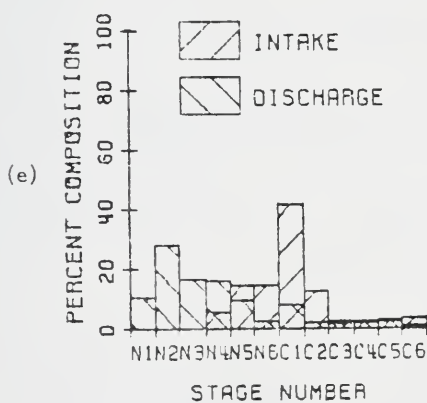
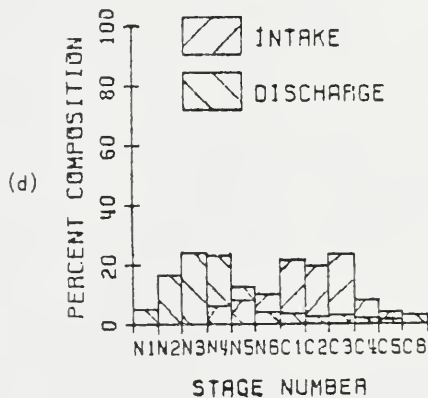
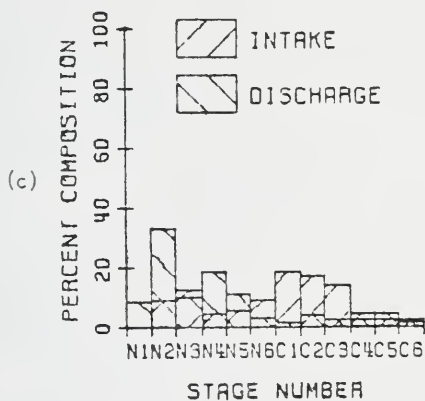
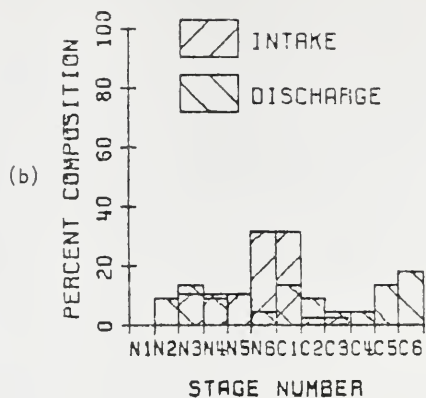
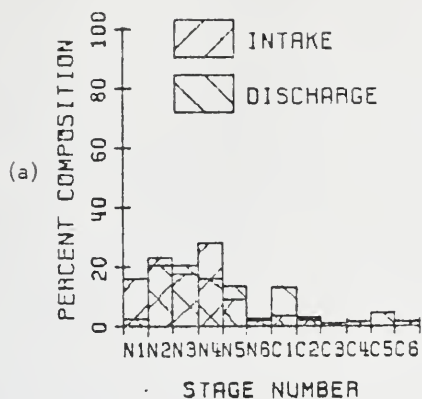


Fig. B-26. Continued
(g) Day 7, (h) Day 8.

Fig. 8-27. Histogram series for Oithona brevicornis in growth experiment started on August 6, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 4, (e) Day 5, (f) Day 6.



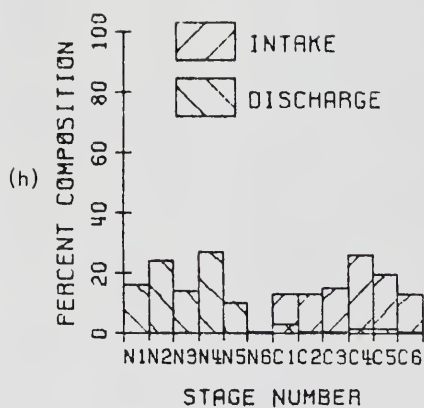
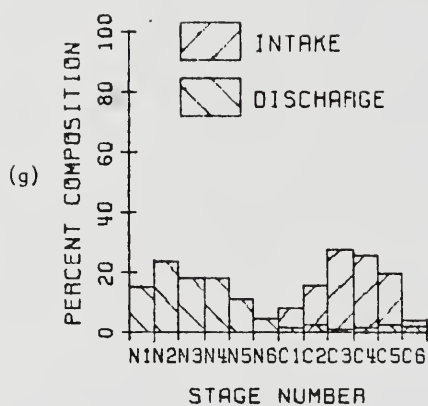
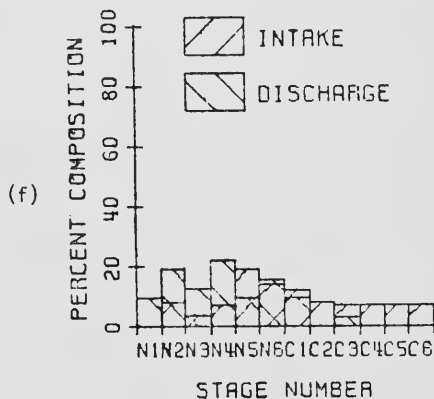
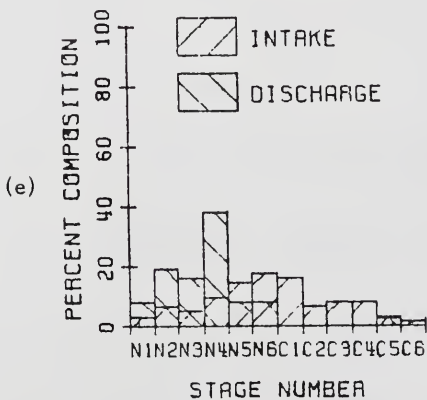
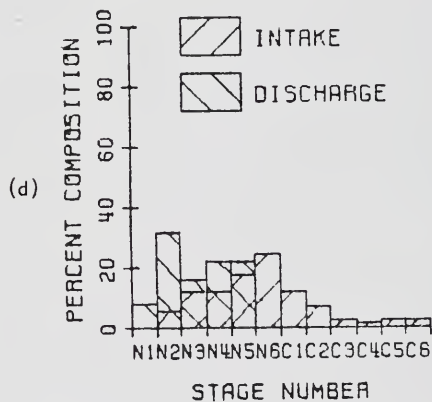
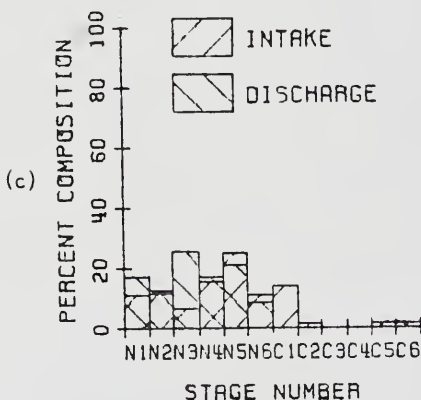
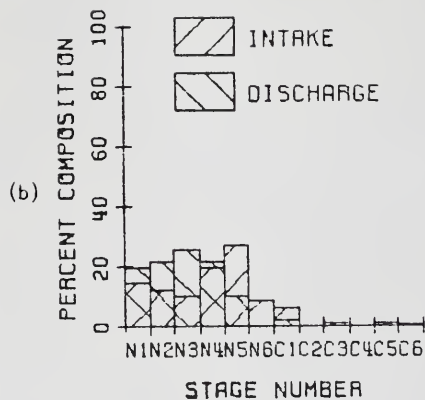
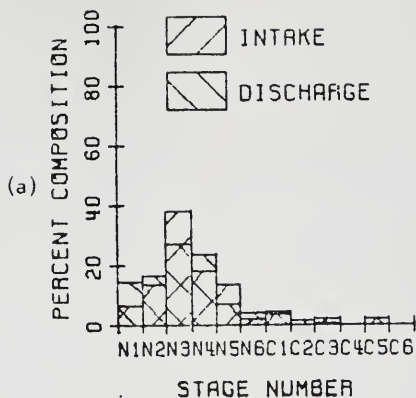


Fig. B-27. Continued
(g) Day 7, (h) Day 8.

Fig. B-28. Histogram series for Oithona brevicornis in growth experiment started on August 20, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 2, (c) Day 3, (d) Day 4, (e) Day 5, (f) Day 6.



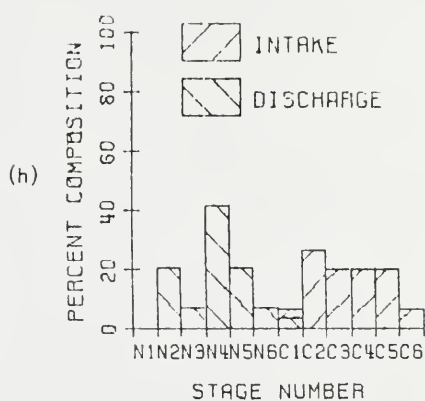
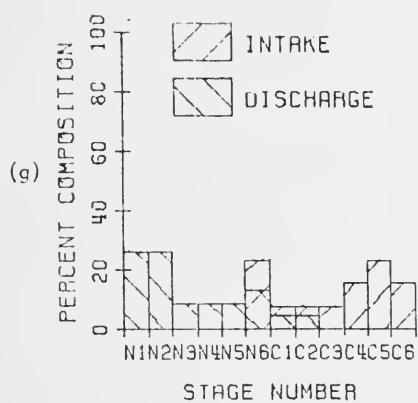
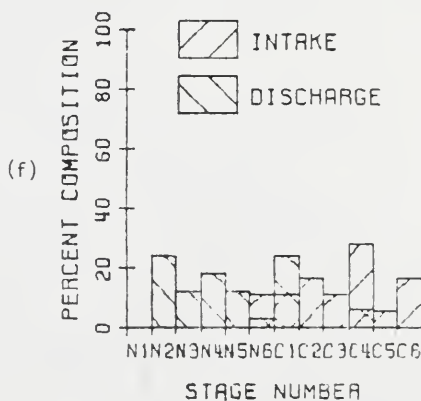
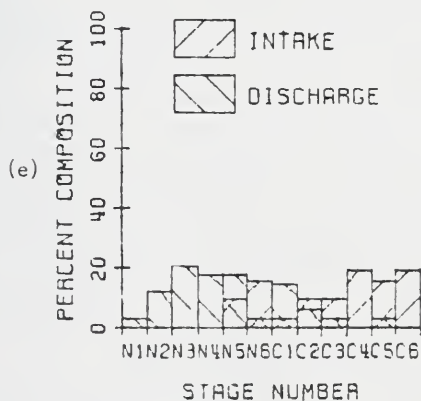
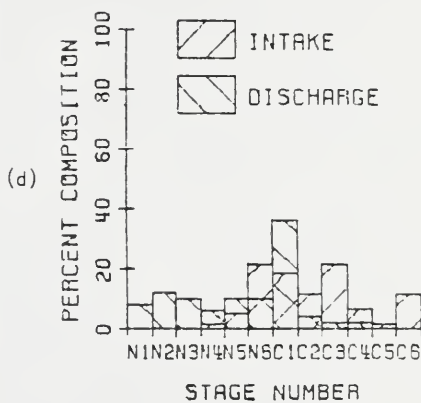
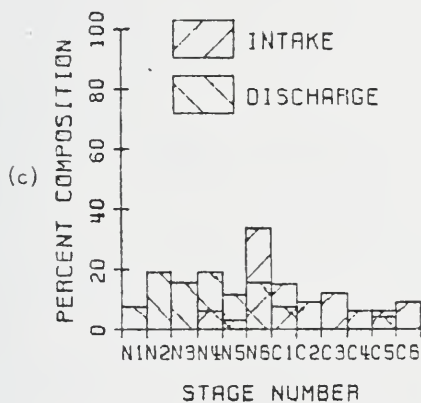
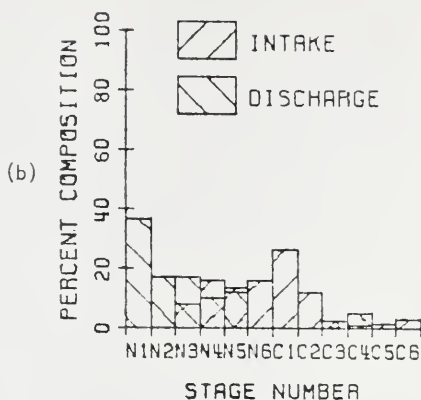
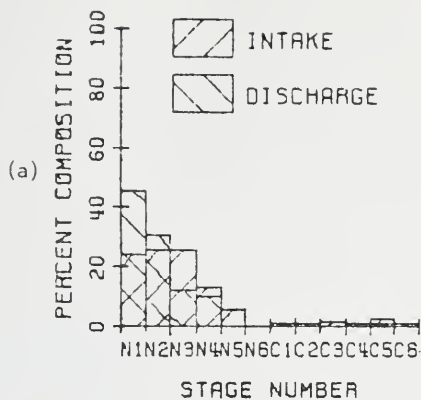


Fig. B-28. Continued
(g) Day 7, (h) Day 8.

Fig. B-29. Histogram series for Oithona brevicornis in growth experiment started on September 3, 1974, showing changes in population structure with time: (a) Day 0.3, (b) Day 3, (c) Day 4, (d) Day 5, (e) Day 6, (f) Day 7.



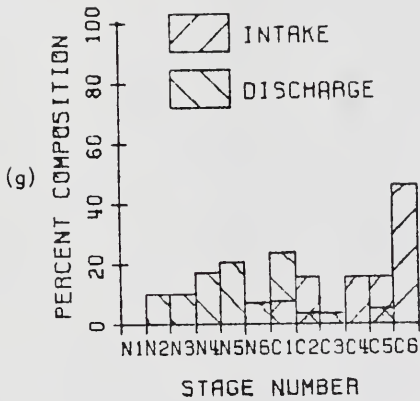


Fig. B-29. Continued
(g) Day 8.

APPENDIX C

Survivorship Curves

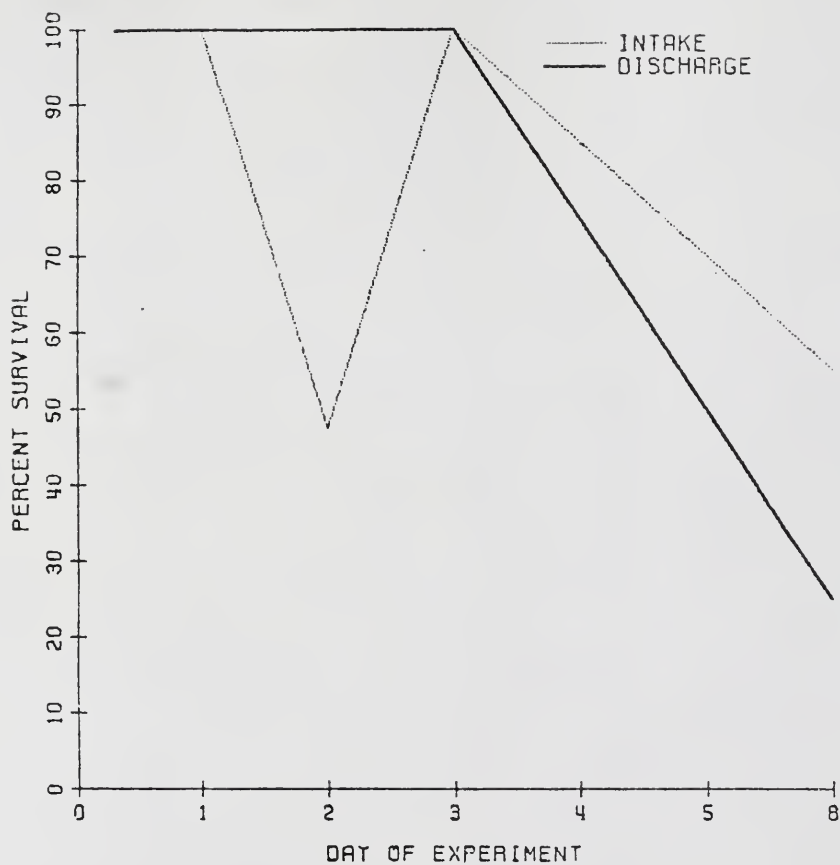


Fig. C-1. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on November 13, 1973. Percent of estimated initial population surviving vs. day of growth experiment.

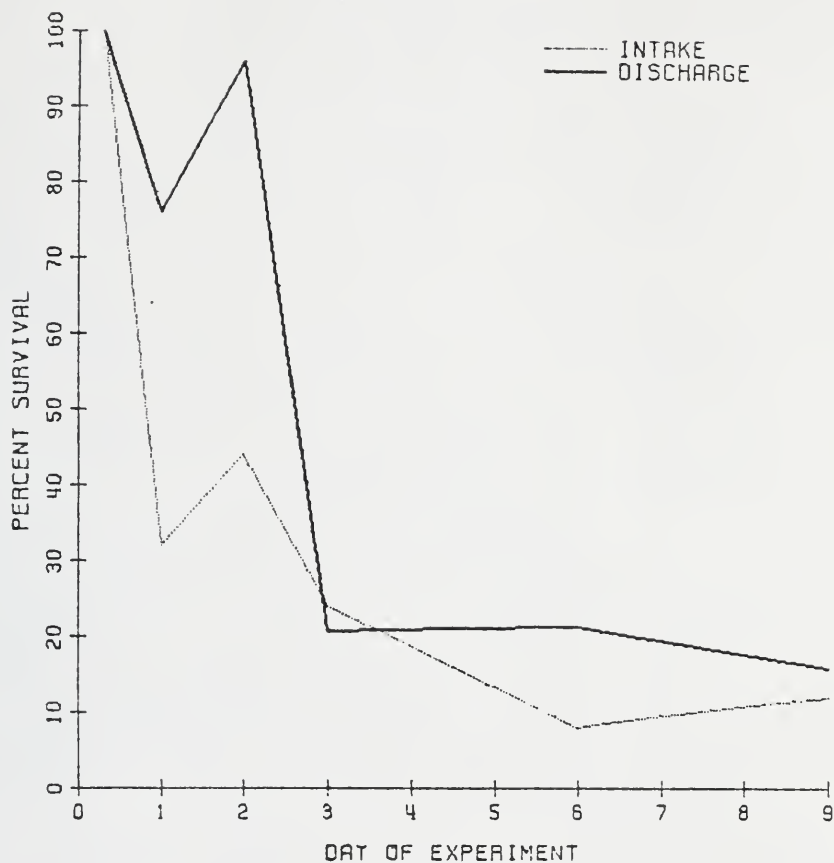


Fig. C-2. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on November 27, 1973. Percent of estimated initial population surviving vs. day of growth experiment.

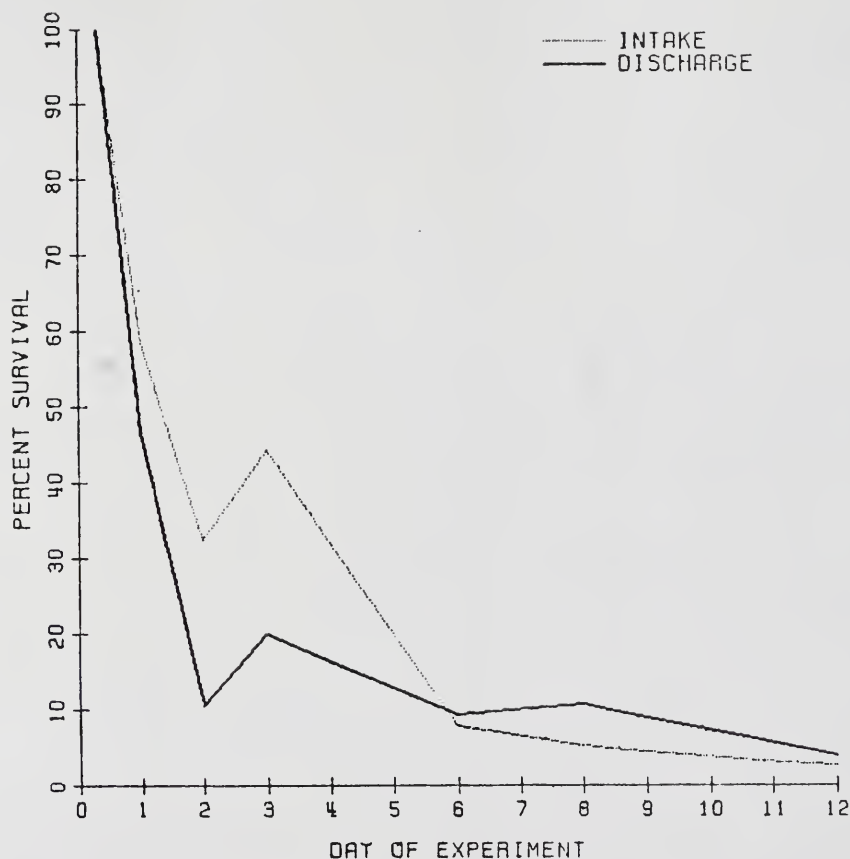


Fig. C-3. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on December 26, 1973. Percent of estimated initial population surviving vs. day of growth experiment.

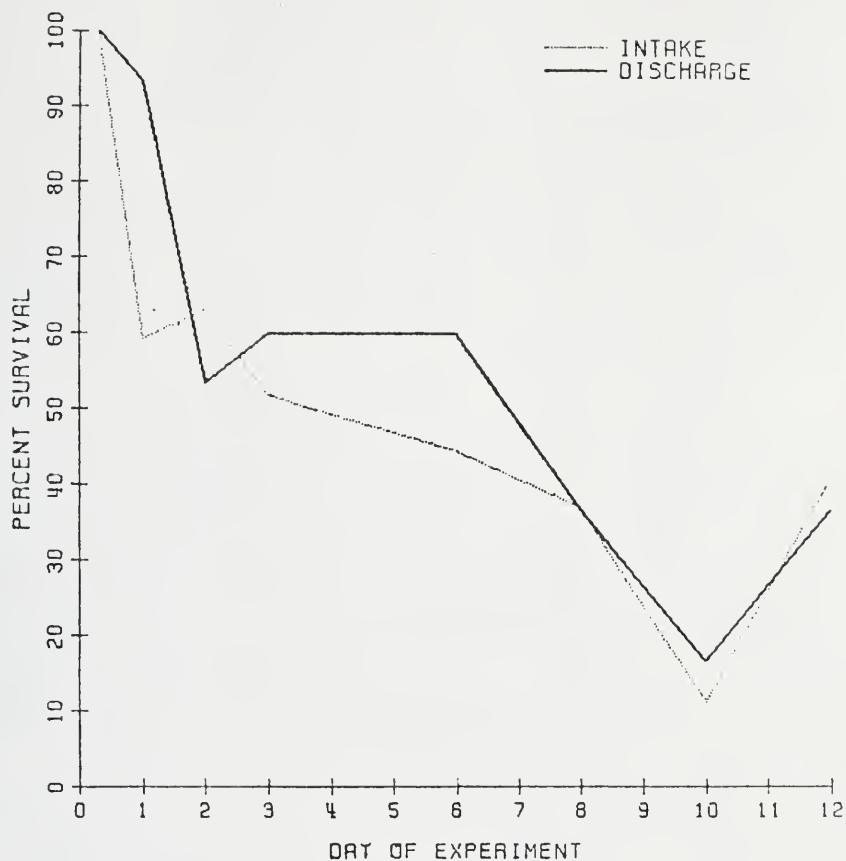


Fig. C-4. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on January 22, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

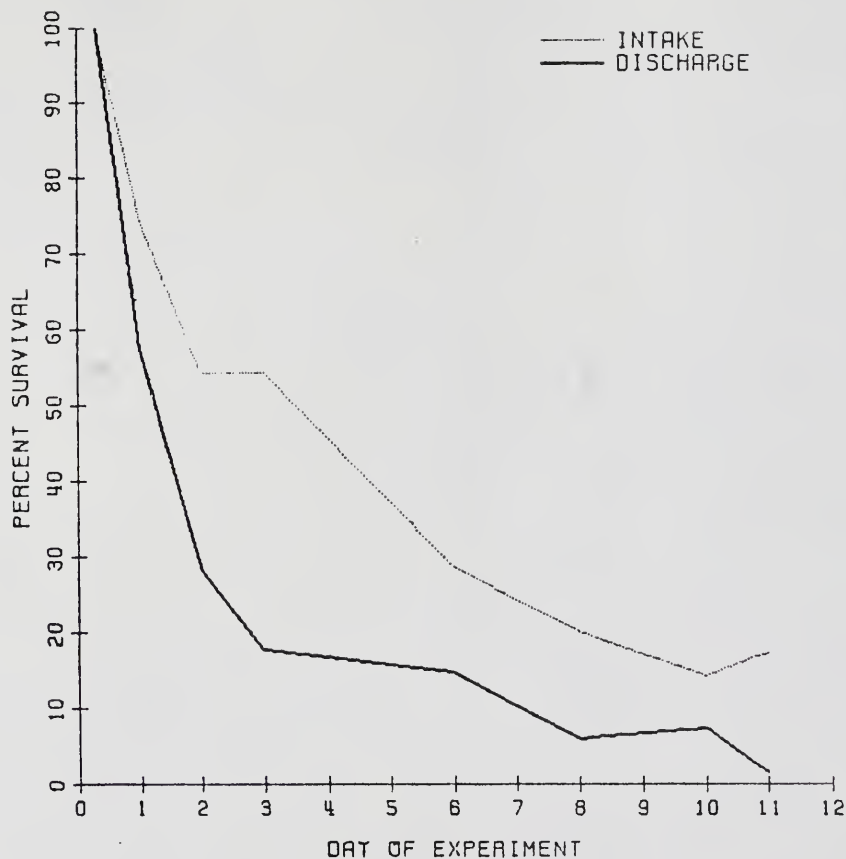


Fig. C-5. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on February 5, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

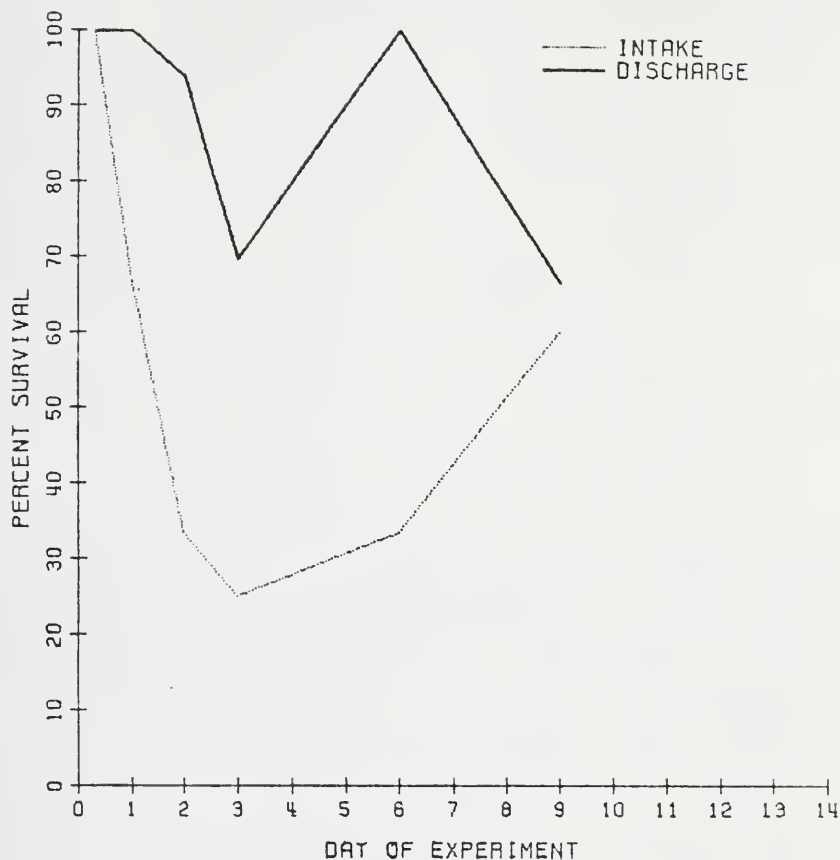


Fig. C-6. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on March 19, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

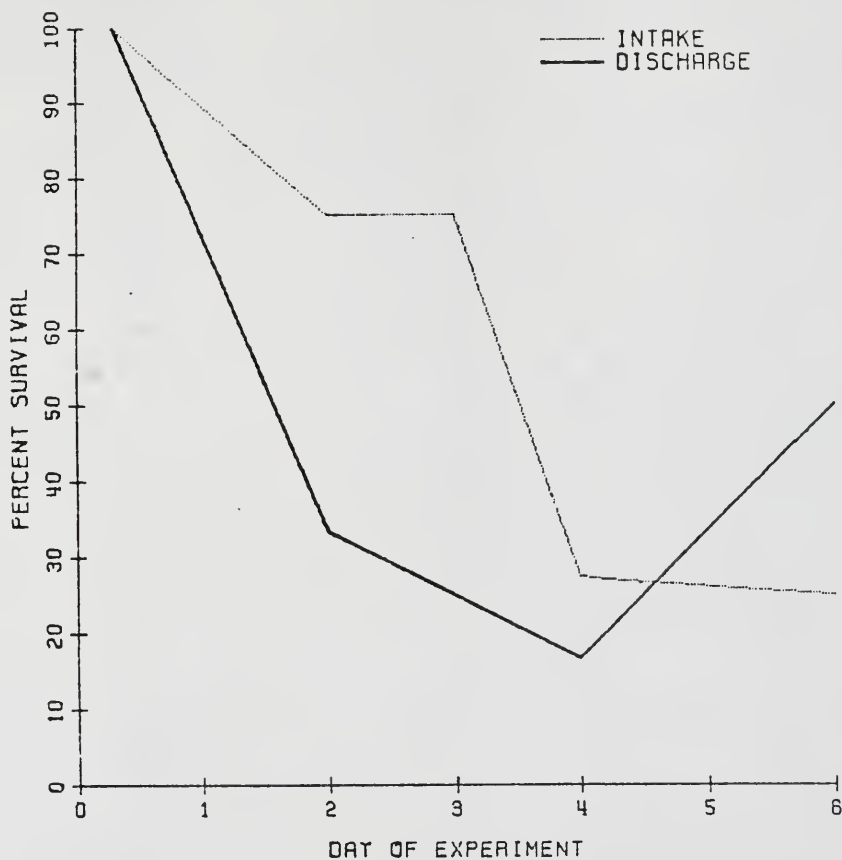


Fig. C-7. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on May 28, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

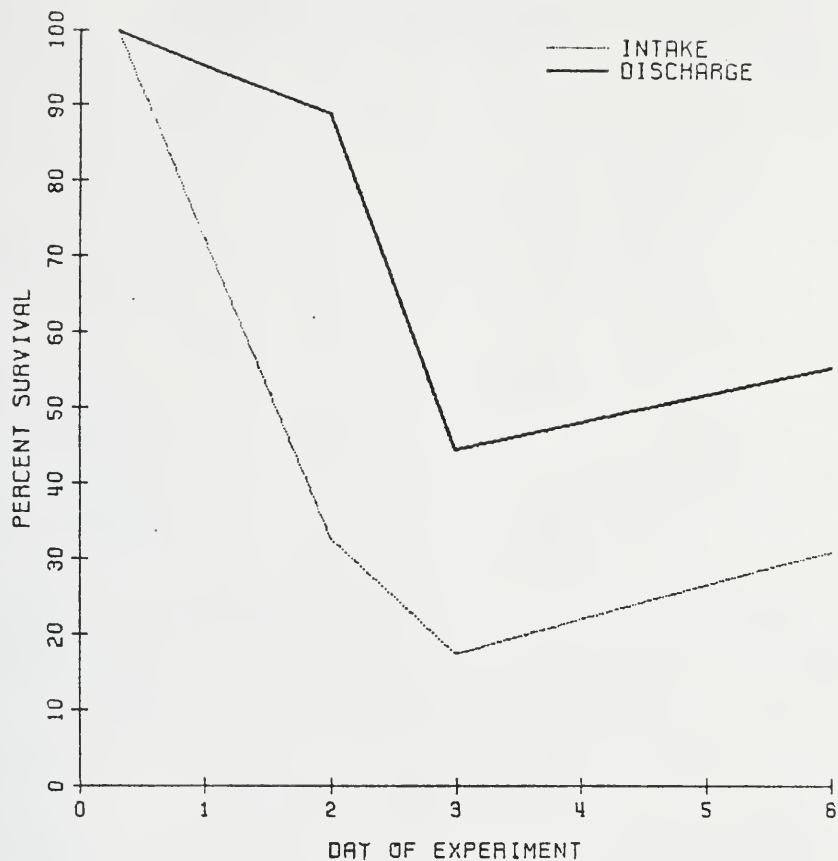


Fig. C-8. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on June 11, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

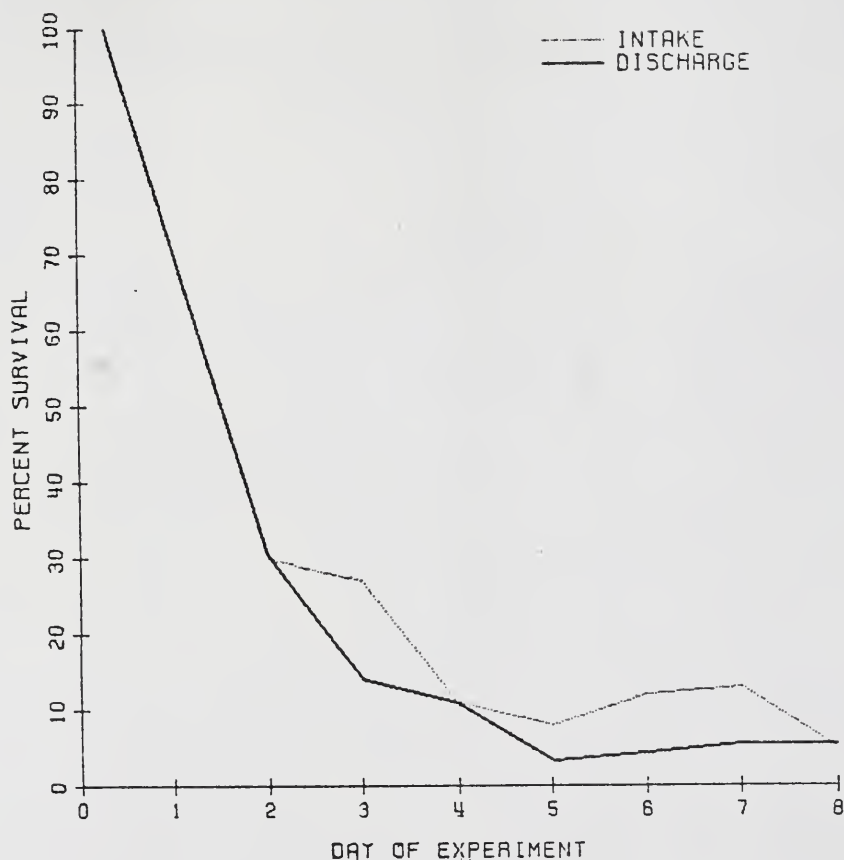


Fig. C-9. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected June 25, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

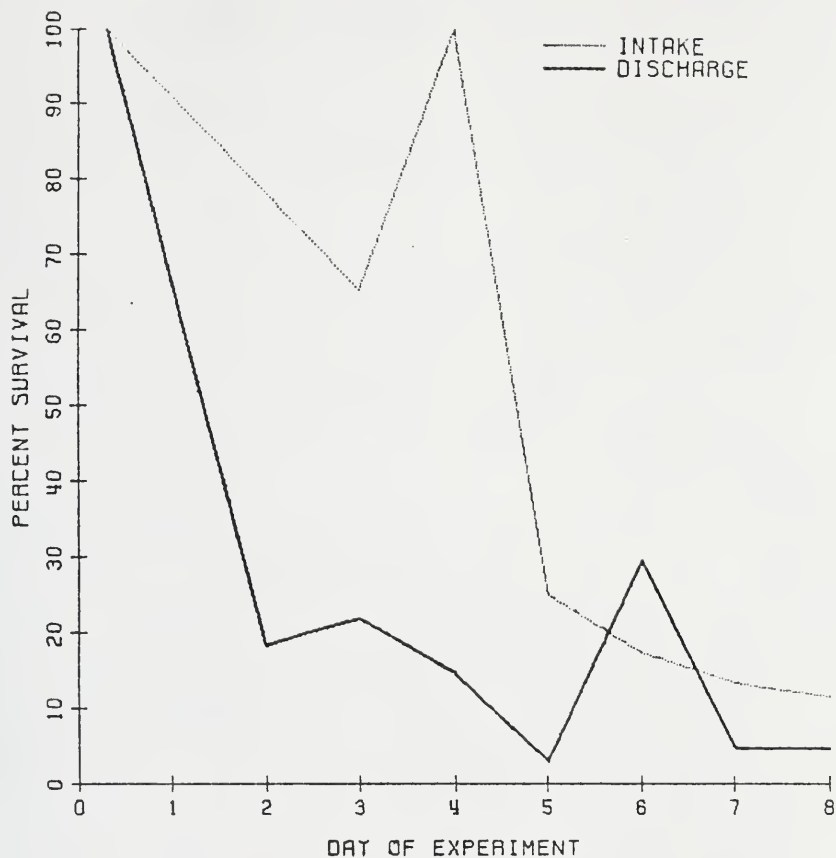


Fig. C-10. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on July 9, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

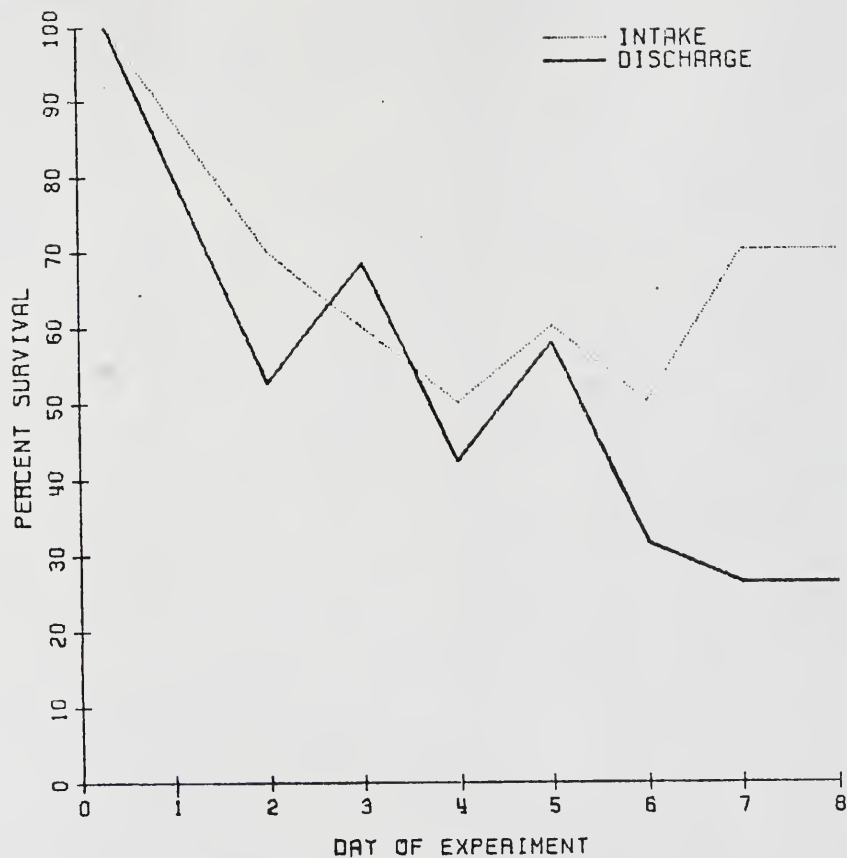


Fig. C-II. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on July 23, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

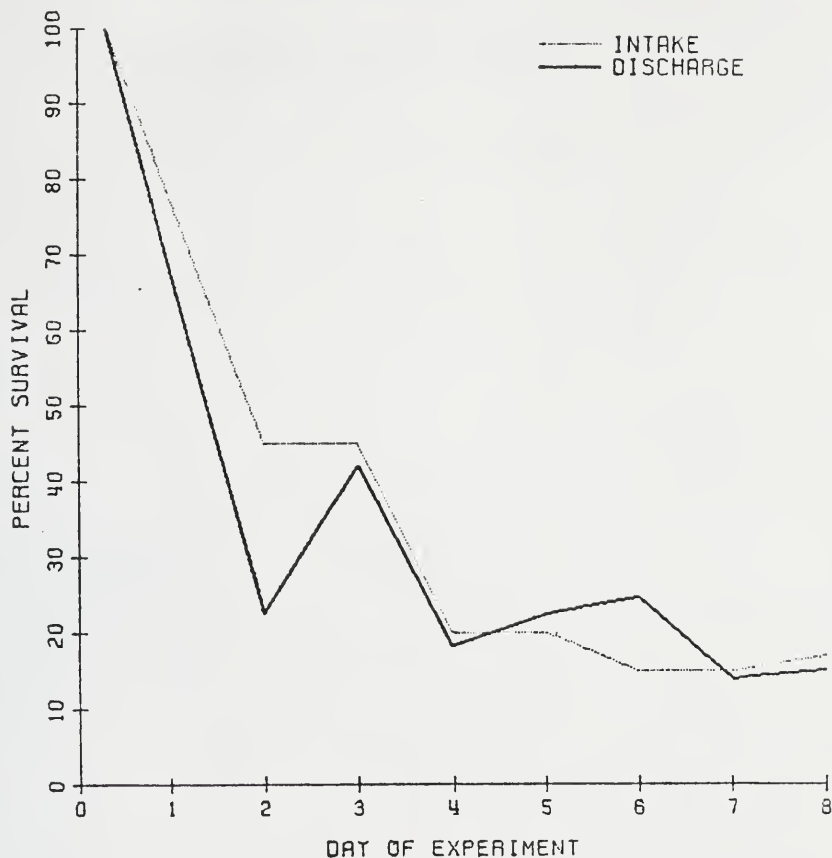


Fig. C-12. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on August 6, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

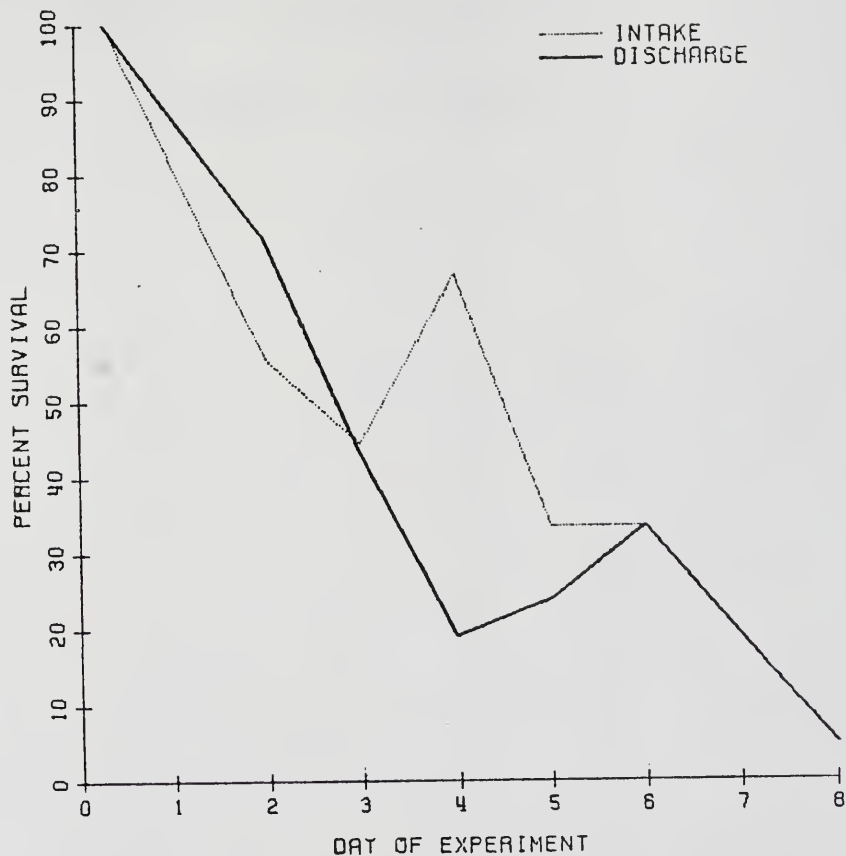


Fig. C-13. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on August 20, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

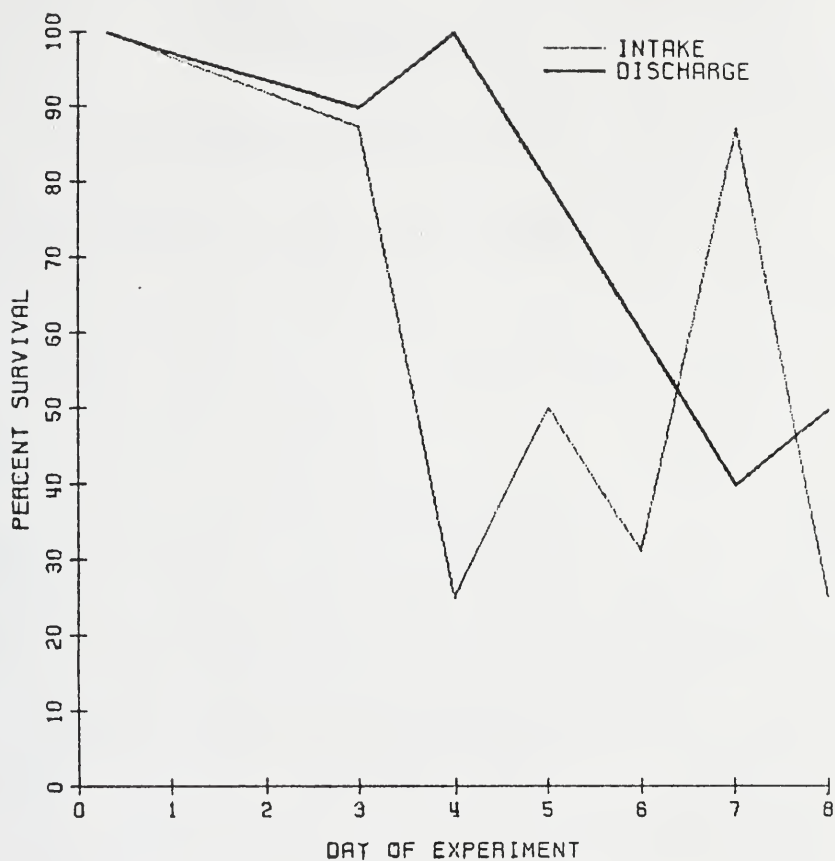


Fig. C-14. Survivorship curve for intake and discharge populations of *Acartia tonsa* juveniles collected on September 3, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

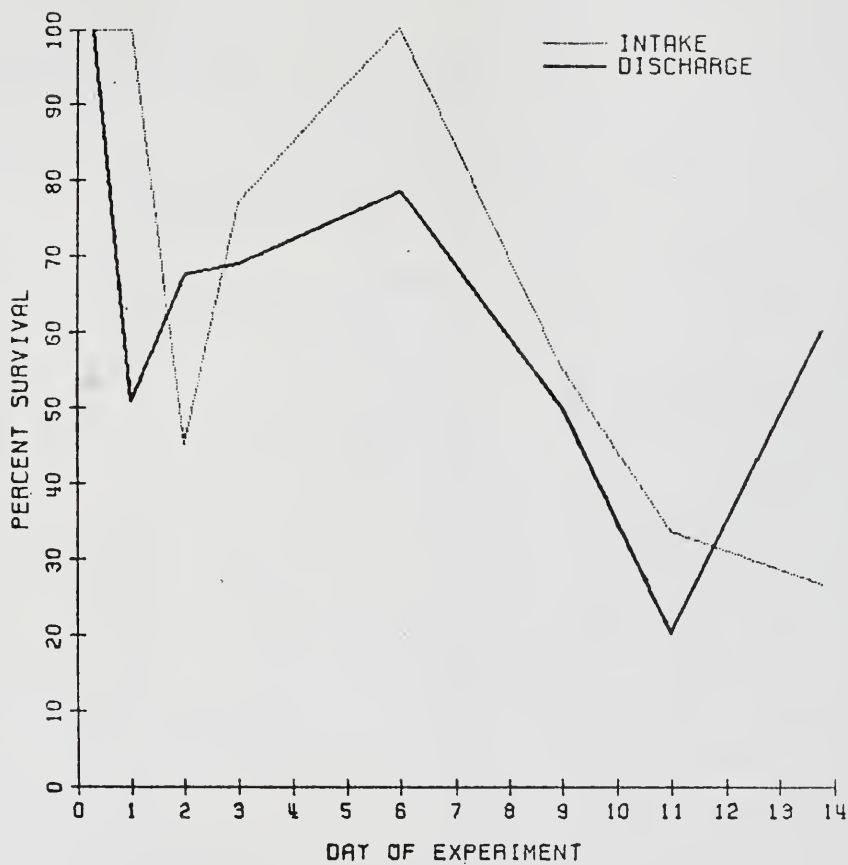


Fig. C-15. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on November 27, 1973. Percent of estimated initial population surviving vs. day of growth experiment.

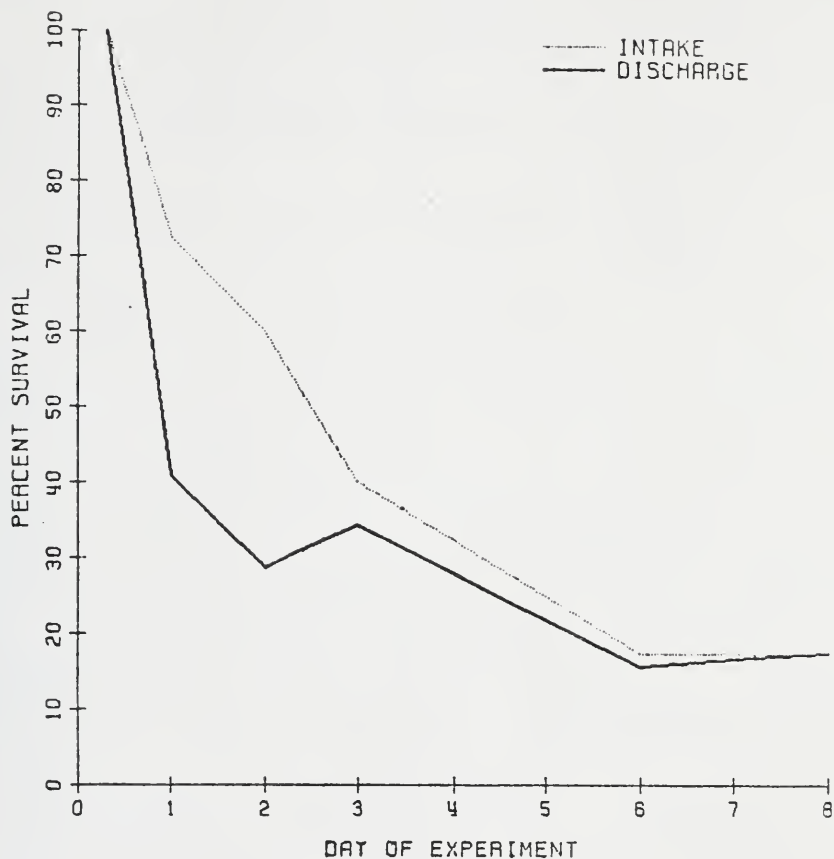


Fig. C-16. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on December 26, 1973. Percent of estimated initial population surviving vs. day of growth experiment.

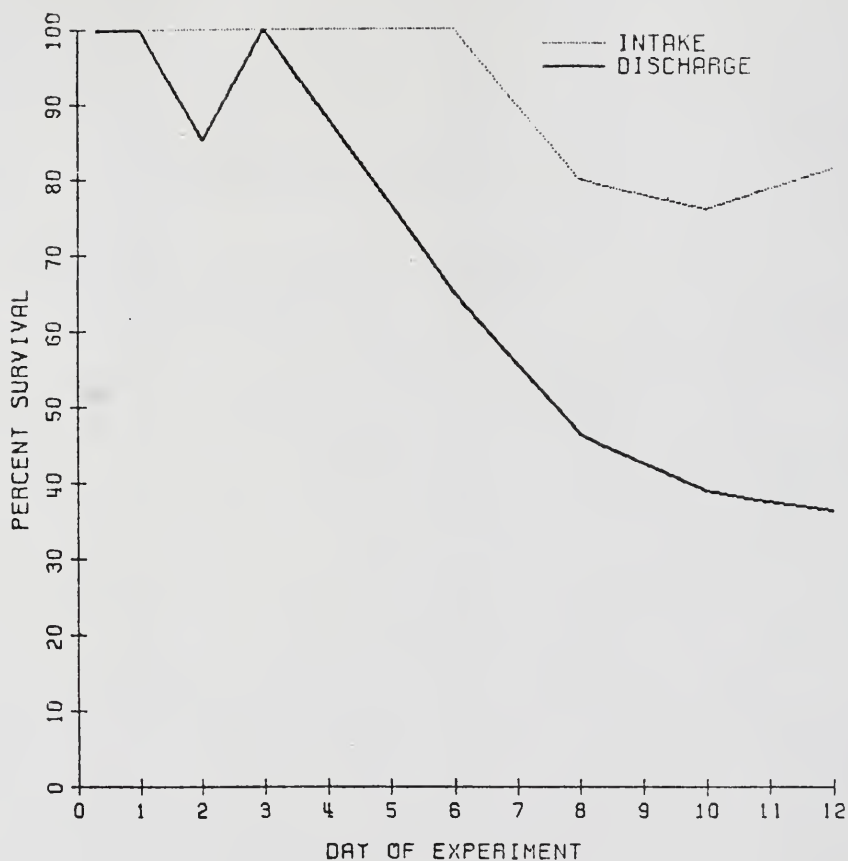


Fig. C-17. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on January 22, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

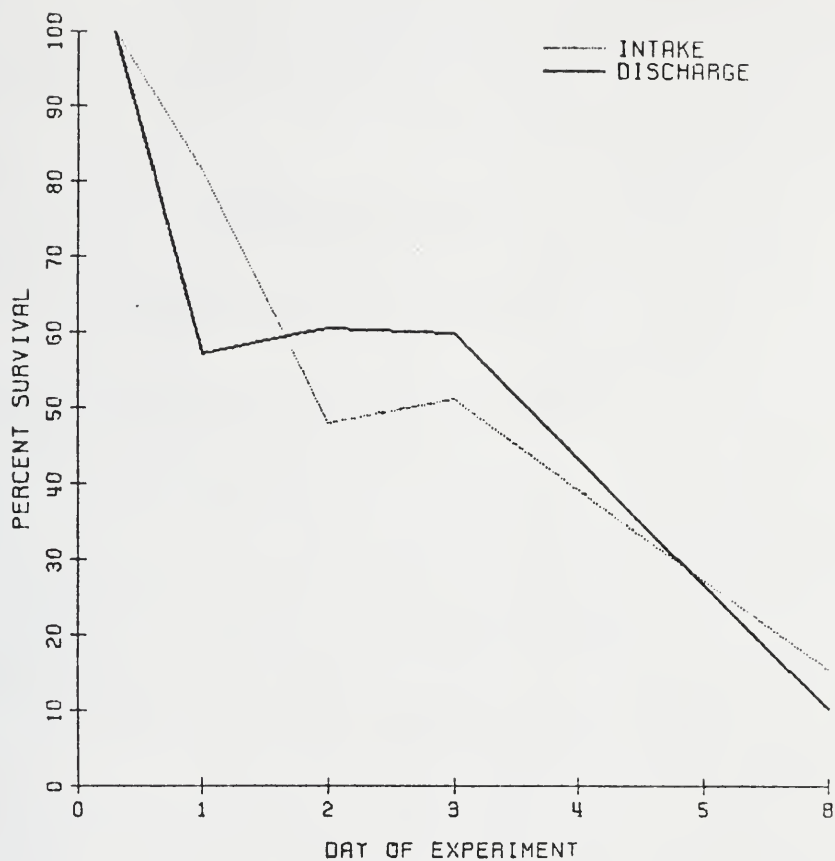


Fig. C-18. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on February 5, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

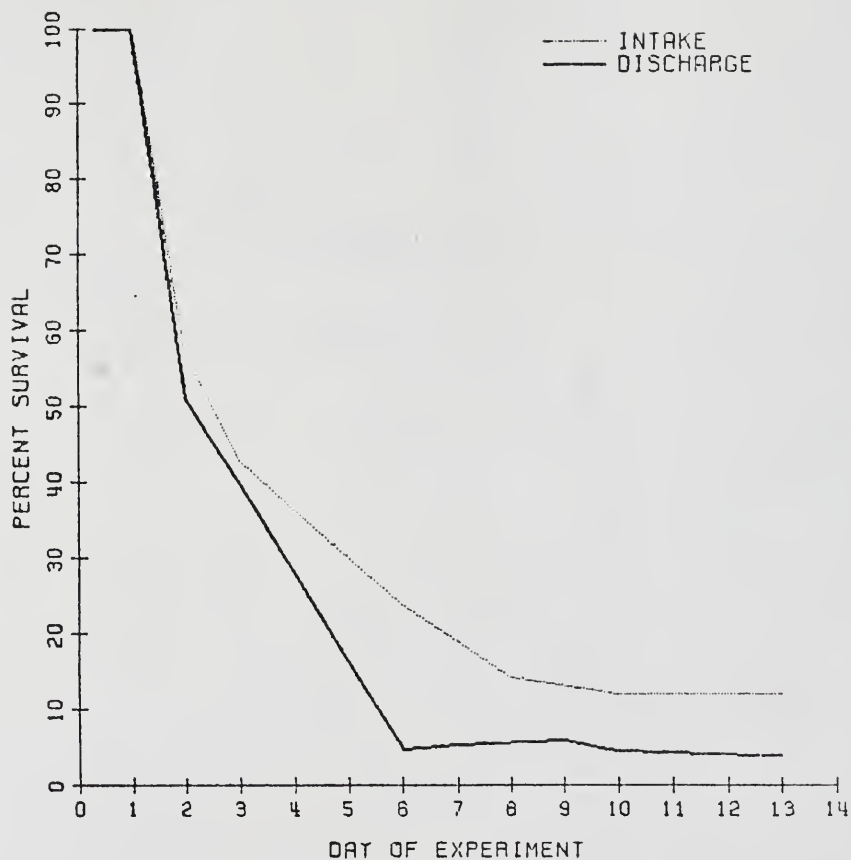


Fig. C-19. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on March 19, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

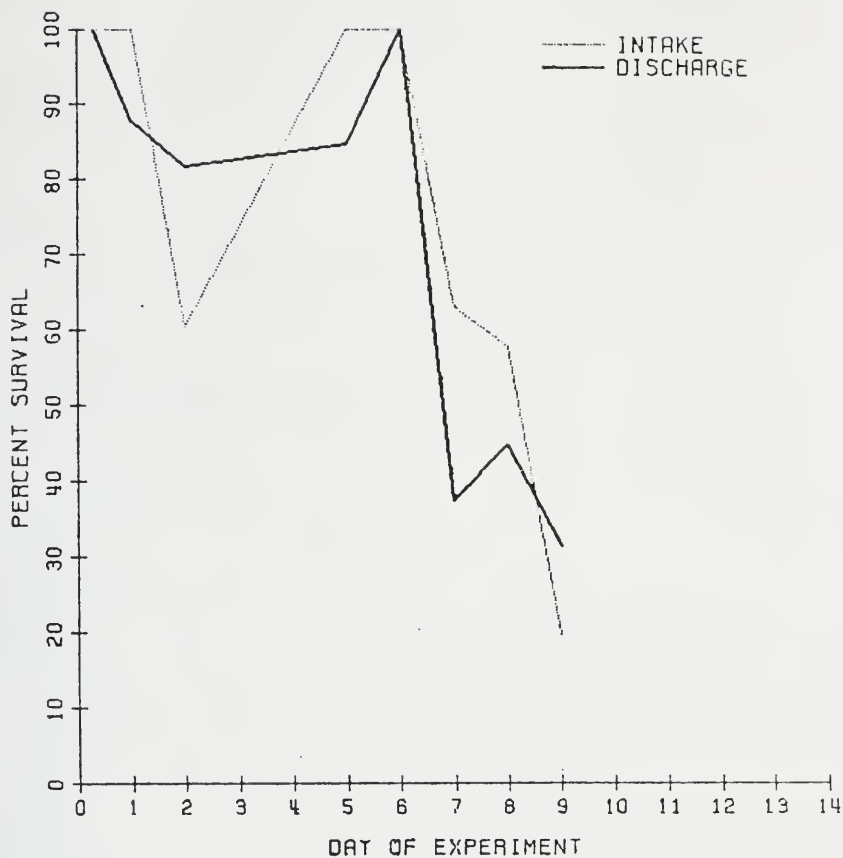


Fig. C-20. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on April 24, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

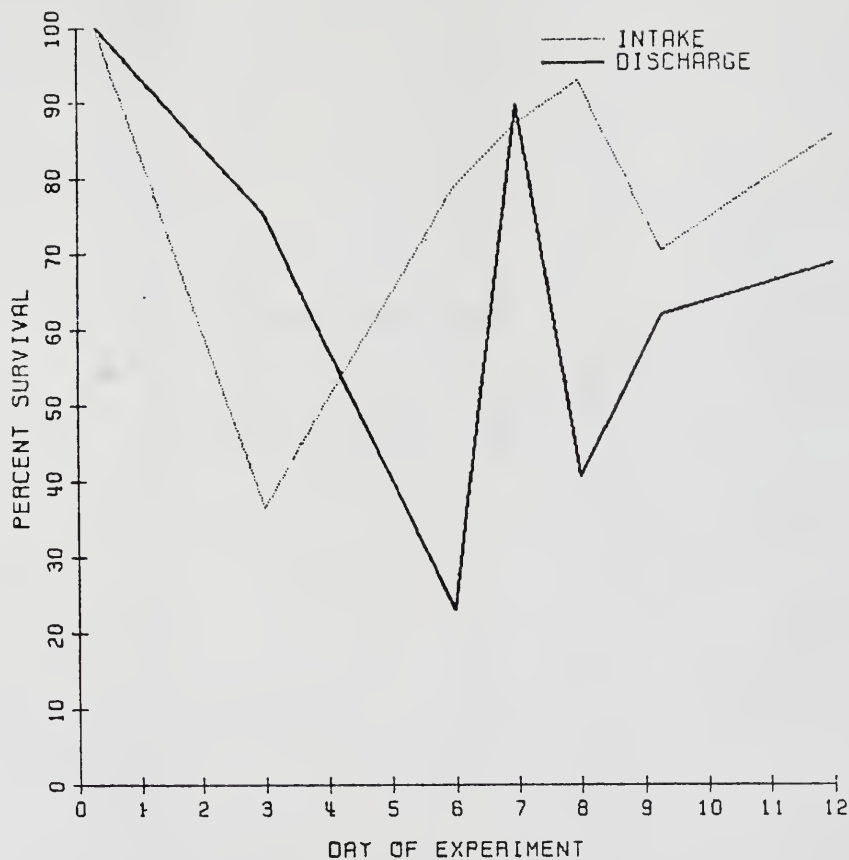


Fig. C-21. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on May 14, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

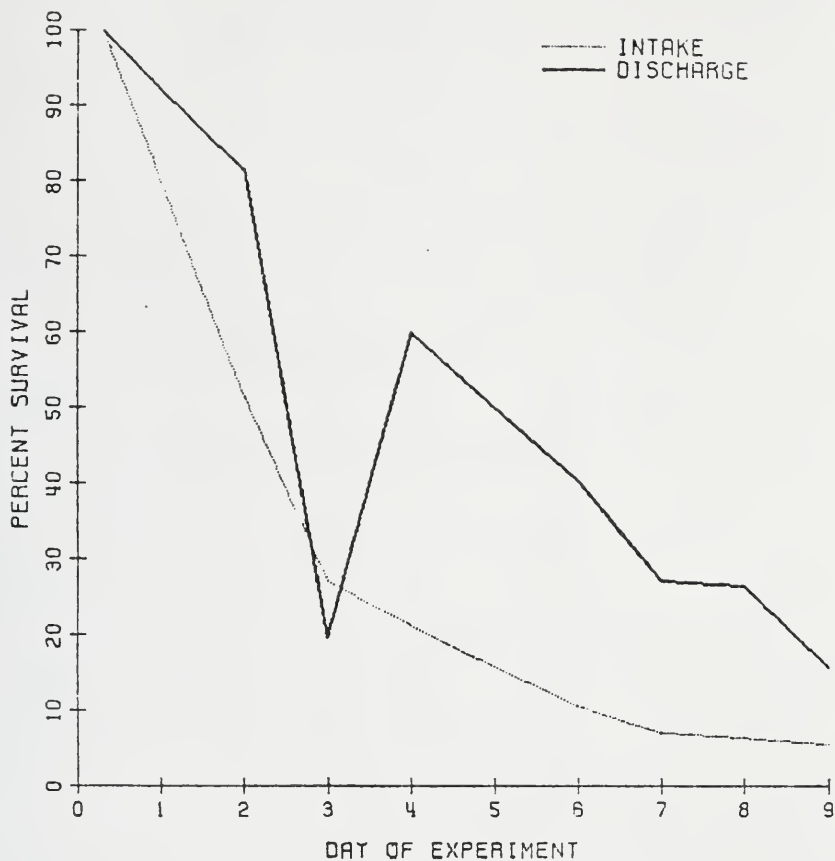


Fig. C-22. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on May 28, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

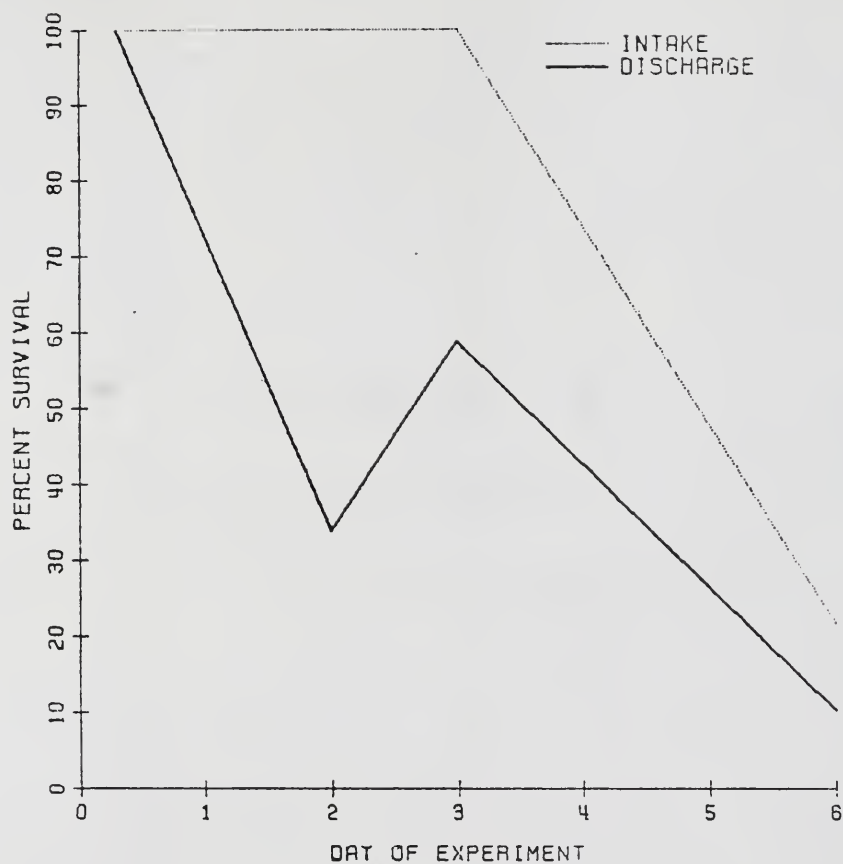


Fig. C-23. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on June 11, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

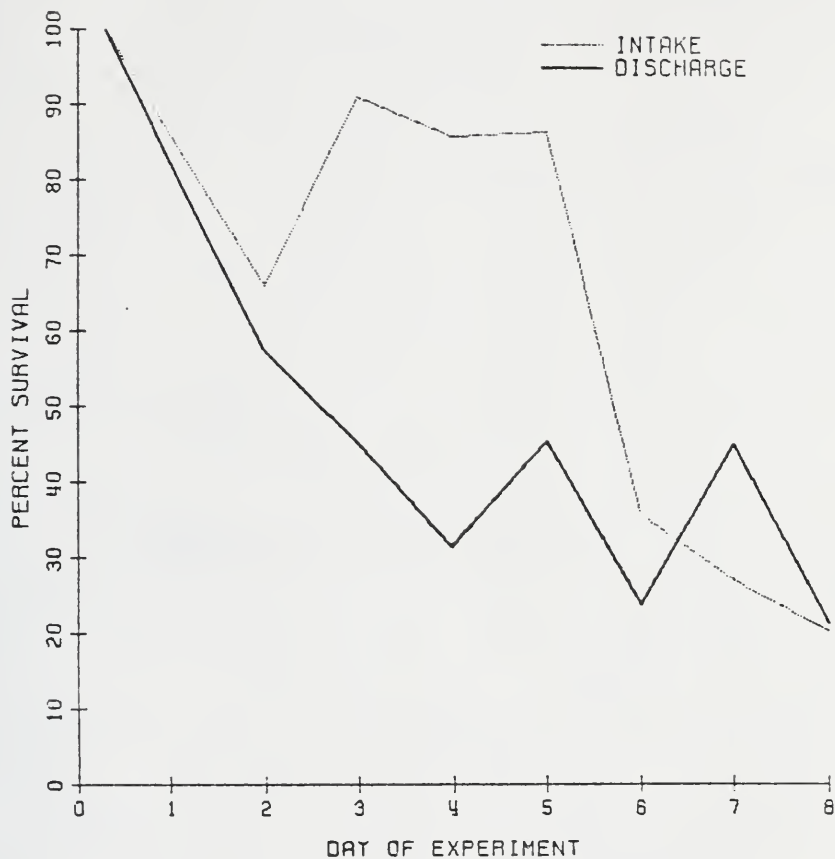


Fig. C-24. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on June 25, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

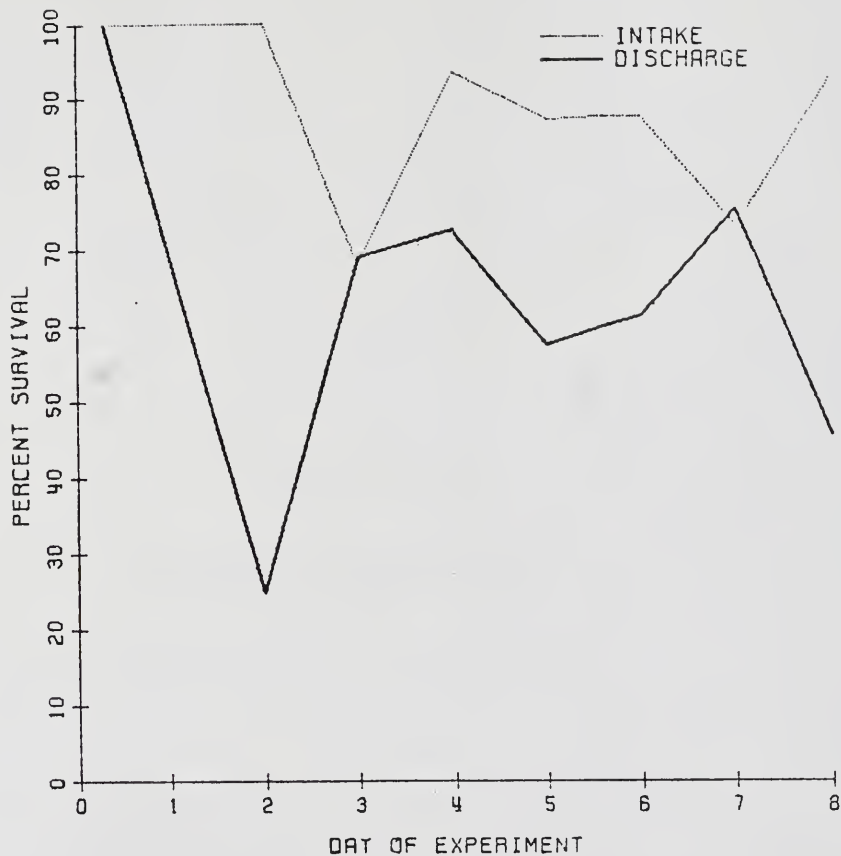


Fig. C-25. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on July 9, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

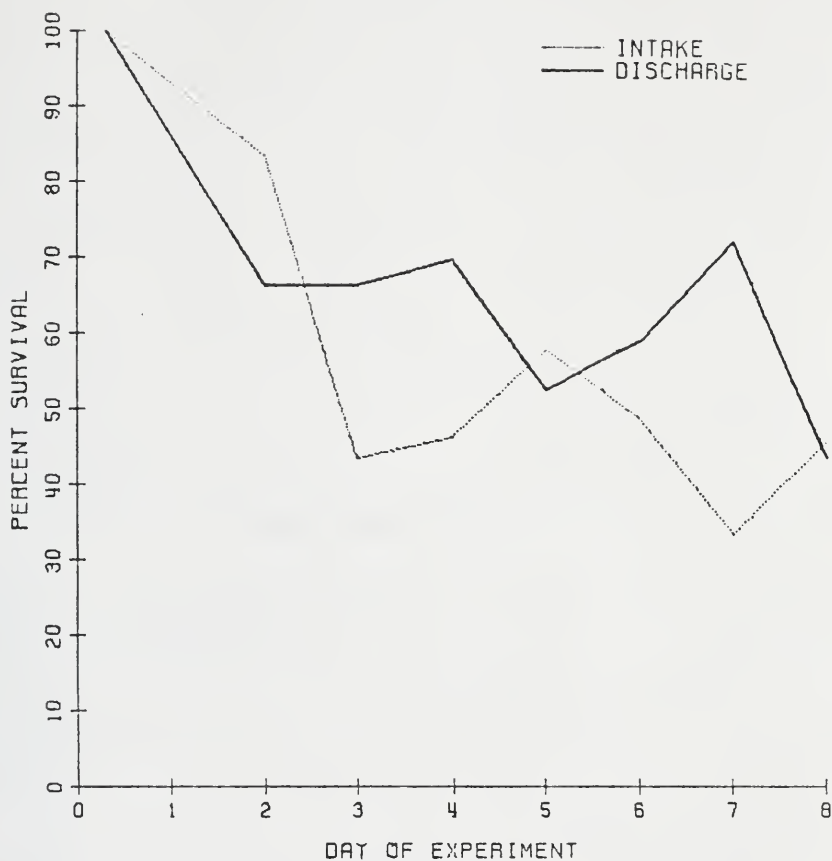


Fig. C-26. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on July 23, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

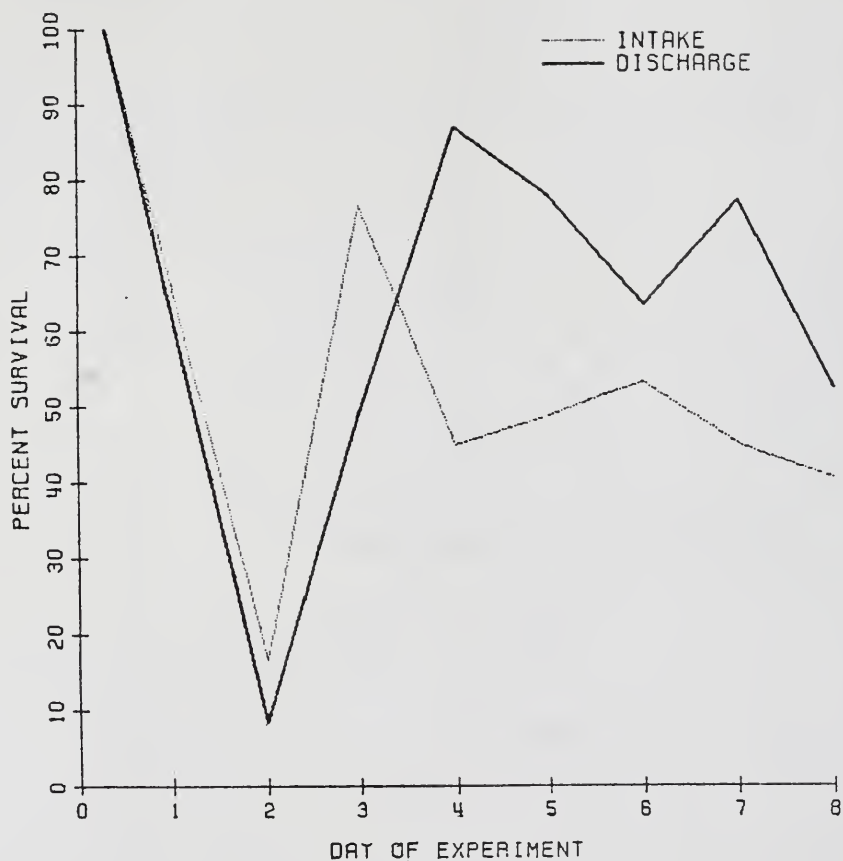


Fig. C-27. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on August 6, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

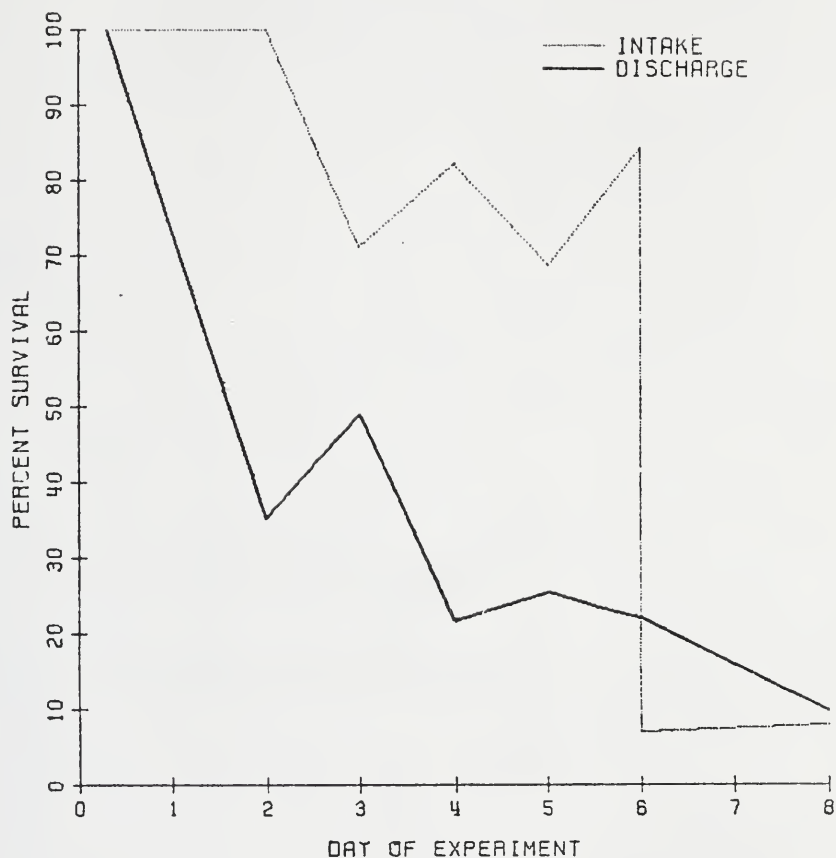


Fig. C-28. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on August 20, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

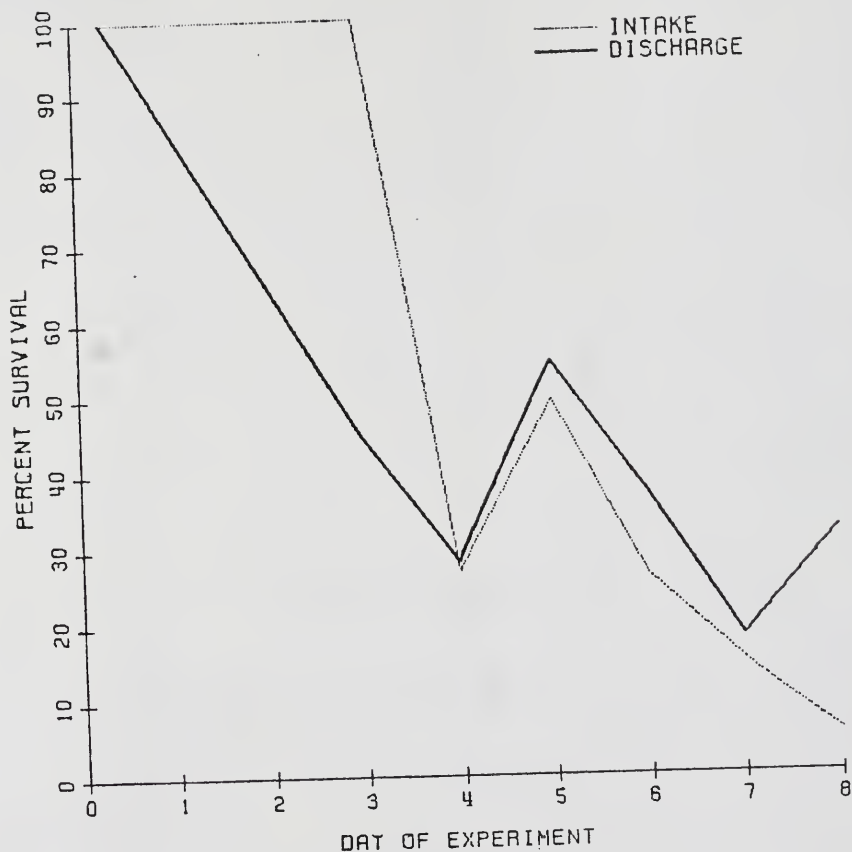


Fig. C-29. Survivorship curve for intake and discharge populations of *Oithona brevicornis* juveniles collected on September 3, 1974. Percent of estimated initial population surviving vs. day of growth experiment.

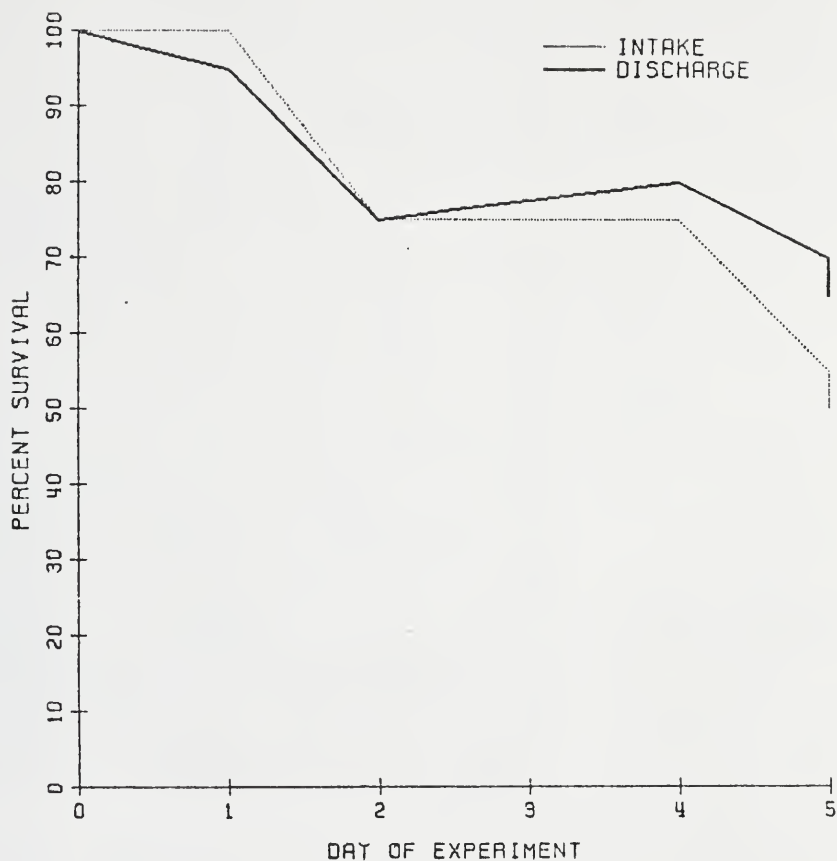


Fig. C-30. Survivorship curve for intake and discharge populations of *Acartia tonsa* adults collected on April 2, 1974. Percent of initial population surviving vs. day of reproductive experiment.

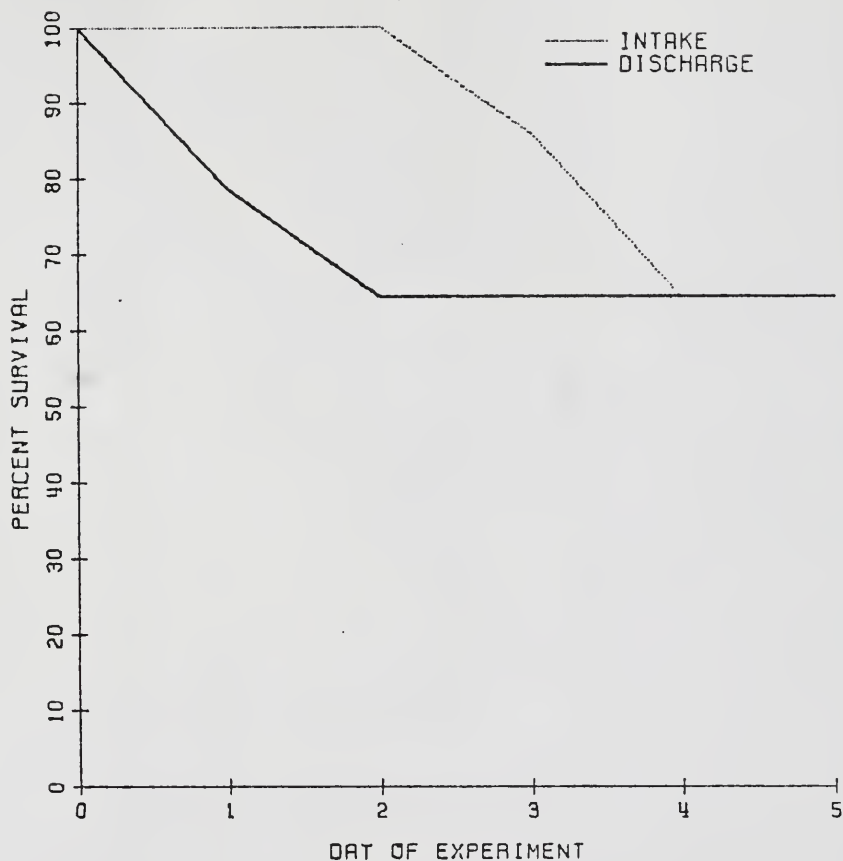


Fig. C-31. Survivorship curve for intake and discharge populations of *Acartia tonsa* adults collected on May 14, 1974. Percent of initial population surviving vs. day of reproductive experiment.

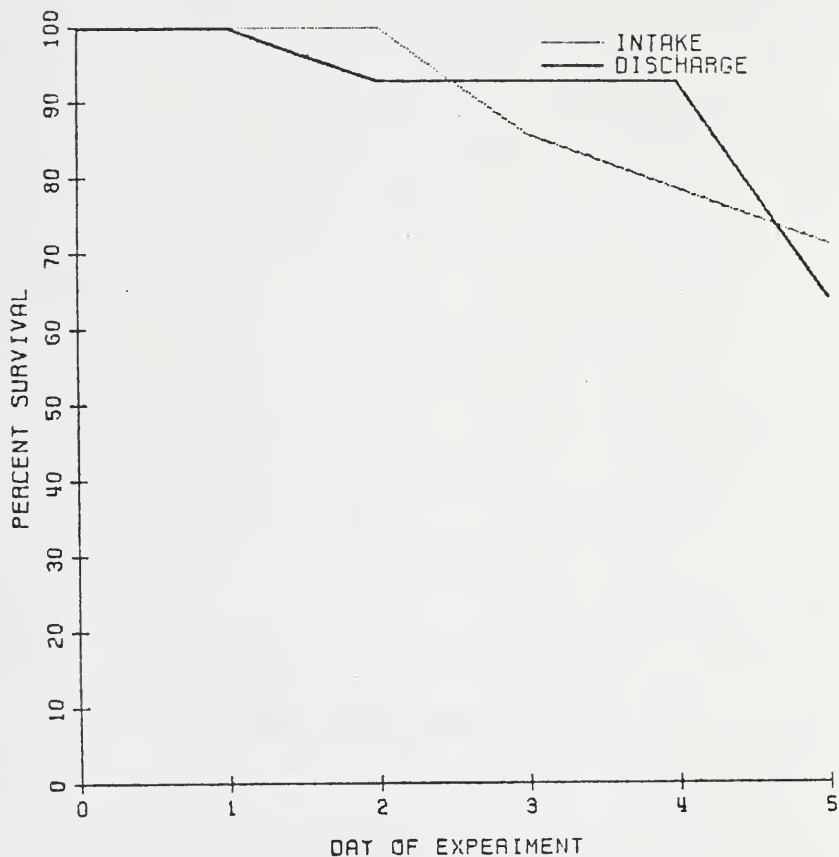


Fig. C-32. Survivorship curve for intake and discharge populations of *Acartia tonsa* adults collected on May 28, 1974. Percent of initial population surviving vs. day of reproductive experiment.

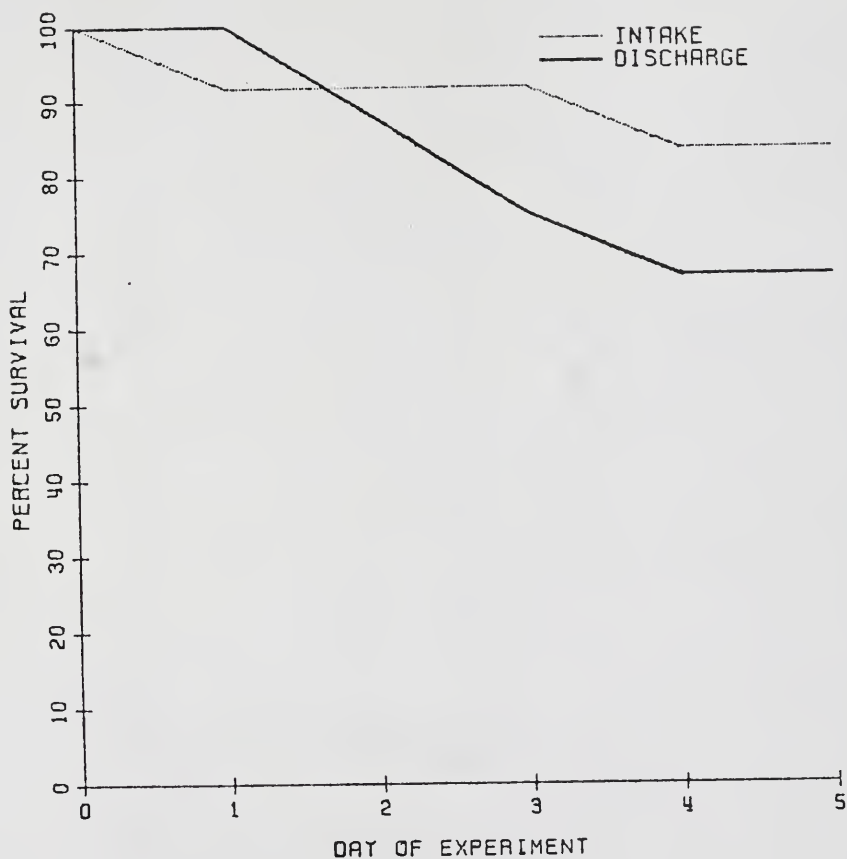


Fig. C-33. Survivorship curve for intake and discharge populations of *Acartia tonsa* adults collected on June 11, 1974. Percent of initial population surviving vs. day of reproductive experiment.

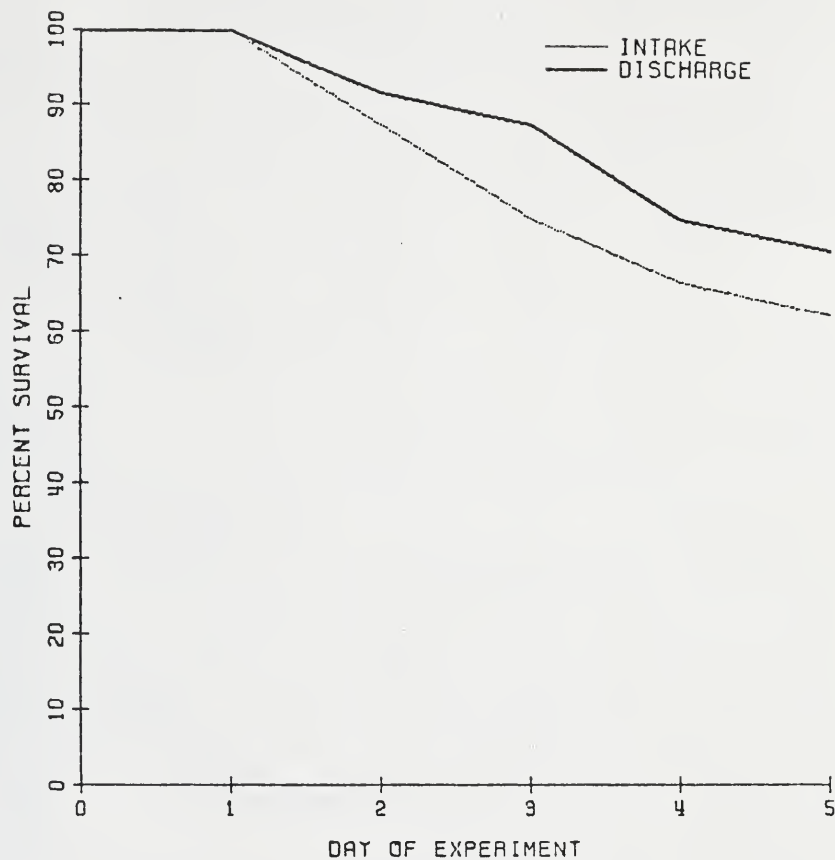


Fig. C-34. Survivorship curve for intake and discharge populations of *Acartia tonsa* adults collected on June 25, 1974. Percent of initial population surviving vs. day of reproductive experiment.

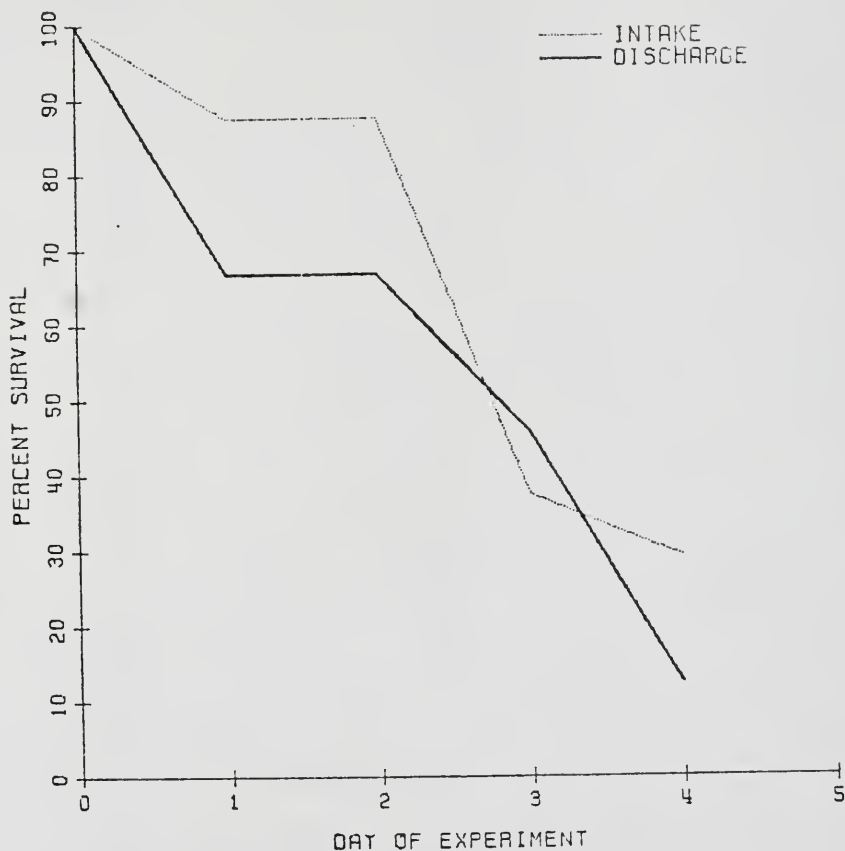


Fig. C-35. Survivorship curve for intake and discharge populations of *Acartia tonsa* adults collected on July 9, 1974. Percent of initial population surviving vs. day of reproductive experiment.

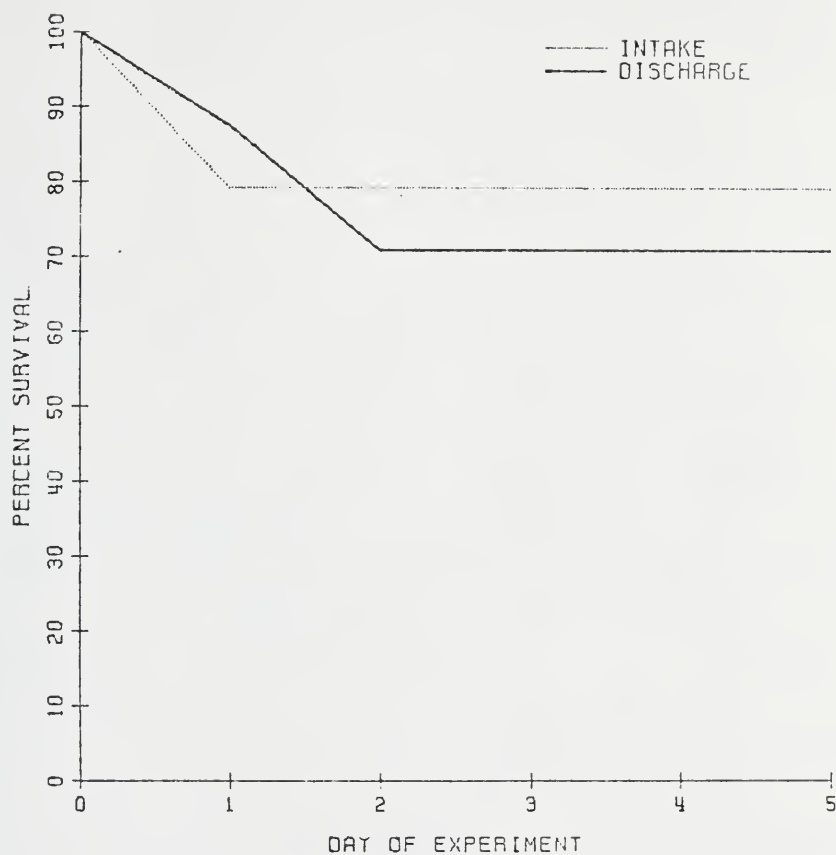


Fig. C-36. Survivorship curve for intake and discharge populations of *Acartia tonsa* adults collected on July 23, 1974. Percent of initial population surviving vs. day of reproductive experiment.

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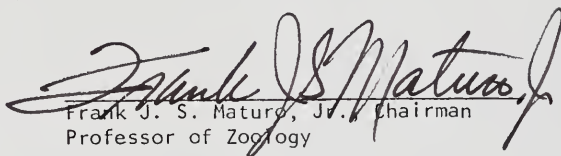
BIOGRAPHICAL SKETCH

Raymond William Alden III was born December 29, 1949, in Daytona Beach, Florida. In June 1967 he graduated from Newton South High School in Newton, Massachusetts. In June 1971 he received the degree of Bachelor of Arts from Stetson University in DeLand, Florida, graduating magna cum laude. As an undergraduate, he was a member of Beta Beta Beta, an honorary biological society, and Scroll and Key, a Phi Beta Kappa colony.

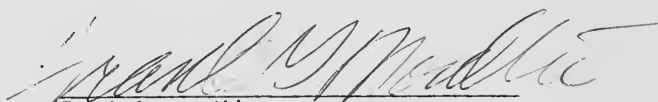
From September 1971 until the present time, Raymond William Alden III has been enrolled in the Graduate School of the University of Florida, majoring in zoology. Having received approval from the Department of Zoology to bypass the Master of Science degree, he has been pursuing the degree of Doctor of Philosophy. Since September 1975 he has been working as a Visiting Research Associate with the Marine Sciences Program of the University of North Carolina, Chapel Hill, North Carolina, while completing final requirements for the Doctor of Philosophy degree at the University of Florida.

Raymond William Alden III is a member of the American Association for the Advancement of Science, the American Society of Zoologists, the Atlantic Estuarine Research Society, the Estuarine Research Federation, the Gulf Estuarine Research Society and the Southeastern Estuarine Research Society.

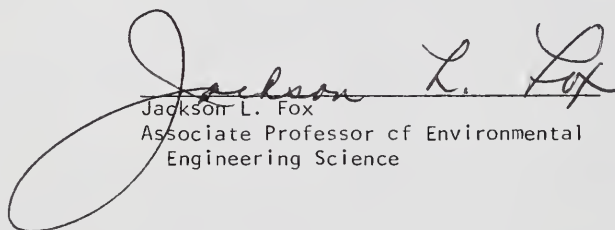
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Frank J. S. Matur, Jr., Chairman
Professor of Zoology

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.


Frank G. Nordlie
Associate Professor of Zoology

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.


Jackson L. Fox
Associate Professor of Environmental
Engineering Science

This dissertation was submitted to the Department of Zoology in the College of Arts and Sciences and to the Graduate Council, and was accepted as partial fulfillment of the requirements for the degree of Doctor of Philosophy.

March 1976

Dean, Graduate School

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